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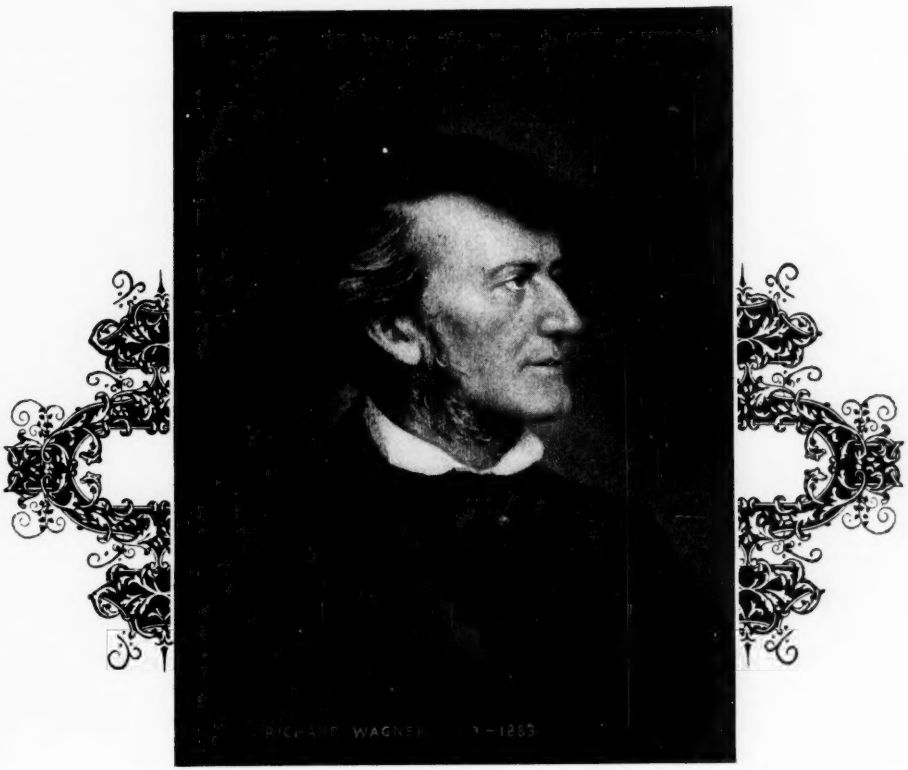

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THE EFFECT OF INTRAVENOUS INFUSIONS OF PHYSIOLOGIC SALINE SOLUTION ON THE PULMONARY ARTERIAL AND PULMONARY CAPILLARY PRESSURE IN MAN [1]

By JOSEPH T. DOYLE,[2] JOSEPH S. WILSON,[3] E. HARVEY ESTES,[4] AND JAMES V. WARREN

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(Submitted for publication November 11, 1950; accepted, January 8, 1951)

In view of the possible role of an increased blood volume in the development of certain features of congestive heart failure, we have undertaken an investigation of experimentally induced changes in plasma volume. The present report deals with the effect of rapid intravenous infusions of physiologic saline solution on the pulmonary arterial and capillary pressure of normal human subjects.

METHODS

Twelve adult men ranging in age from 17 to 64 years and convalescent from noncardiac ailments were studied. The subjects were in the post-absorptive state and in the recumbent position. The pulmonary artery was catheterized by the usual technique (1). An indwelling needle was inserted into a peripheral artery. Expired air was collected for three minutes in a Douglas bag. The oxygen content of the blood samples was determined by the method of Van Slyke and Neill (2) and of the air samples by the method of Haldane (3). The cardiac output was calculated utilizing the Fick principle.

The so-called pulmonary blood volume was determined by the dye dilution method of Stewart and Hamilton (4, 5) as modified by Ebert (6). An accurately measured amount of T-1824 was injected into the cardiac catheter and the arterial blood sampled for two-second periods for 22 seconds after the injection. A ten-minute arterial blood specimen was obtained for estimation of the initial general blood volume. Subsequent blood volumes were calculated from the hematocrit reading, assuming the red blood cell mass to remain constant. The mean pulmonary circulation time and the cardiac output were calculated from the dye time-concentration curve and in turn the pulmonary blood volume was derived.

The instantaneous and mean pulmonary arterial pressure curves were measured from a zero level 5 cm. below the sternal angle by a Sanborn electromanometer and registered with a simultaneous electrocardiographic lead on a Sanborn Polyviso recorder. The so-called pulmonary capillary pressure was measured by the method of Dexter (7). The heart rate was determined from the electrocardiogram.

After completion of these control observations, physiologic saline solution was injected into a peripheral vein at rates of from 77–146 cc. per minute in amounts of 950–1,000 cc. During this time the pulmonary arterial pressure was measured continuously or at frequent intervals. Toward the end of the infusion period the cardiac output and the pulmonary blood volume were again determined. The infusion was then discontinued and in most instances the cardiac output was determined a third time at varying subsequent intervals.

Measurements of the vital capacity were made on eight subjects at various times during the experimental period.

RESULTS

In each of the 12 cases there was a rise in the pulmonary arterial pressure evident within one or two minutes and continuing to increase throughout the infusion period (Table I). The mean resting pulmonary arterial pressure of 7–15 mm. Hg (average 12 mm. Hg) rose to levels of 16–25 mm. Hg (average 20 mm. Hg), representing increases of from 43–150 per cent. Both the systolic and diastolic pressures participated in the rise although the pulse pressure often widened moderately. Within 15 to 60 minutes after the infusion the mean pulmonary arterial pressure had returned to or toward normal.

In four cases tabulated and in four other cases not tabulated here, the rapid infusion of saline solution caused an increase in the pulmonary capillary pressure proportional to that observed in the pulmonary artery. Percentagewise the increase was greater because of the low initial level of the capillary pressure.

Hemodilution at the end of the infusion was indicated by a fall in the hematocrit reading averaging 13 per cent and by an average increase in the general blood volume of 14 per cent. This repre-

[1] This study was supported in part by a grant from the American Heart Association.

[2] Whitehead Foundation Research Fellow, 1949–50.

[3] Whitehead Foundation Research Fellow, 1950–51.

[4] Alfred Stengel Research Fellow of the American College of Physicians.

TABLE I

Hemodynamic effect of rapid intravenous infusion of physiologic saline solution

Patient	Age	Surface area	Remarks	Hematocrit reading	General blood volume	Pulmonary blood volume	Vital capacity	Pulmonary arterial pressure		
								S	D	Mean
	(*years*)	(*sq. m.*)			(*cc./sq. m.*)	(*cc./sq. m.*)	(*l./sq. m.*)	(*mm. Hg*)	(*mm. Hg*)	(*mm. Hg*)
1	39	1.82	Control	42.4	2,430	650	2.19	15	2	7
			1,000 cc. in 9½ min.	37.1	2,780	650	1.93	22	7	16
			38 min. after infusion	40.4	2,550	—	2.16	16	3	10
2	64	1.60	Control	32.3	2,980	—	—	28	6	15
			1,000 cc. in 11 min.	27.6	3,480	650	—	35	15	23
			60 min. after infusion	29.2	3,290	—	—	28	8	17
3	31	1.68	Control	37.7	2,450	480	1.67	20	5	12
			1,000 cc. in 10 min.	31.6	2,920	570	1.43	36	11	22
4	27	1.65	Control	40.0	2,590	420	1.61	22	3	10
			1,000 cc. in 11 min.	34.3	3,020	440	1.64	38	16	25
			35 min. after infusion	33.2	3,120	—	1.45	18	3	8
5	36	1.63	Control	34.7	2,470	660	—	17	5	8
			1,000 cc. in 11 min.	28.4	3,020	800	—	25	10	16
6	17	1.65	Control	47.5	2,750	590	2.24	20	5	10
			1,000 cc. in 13 min.	42.4	3,080	780	2.16	25	10	19
			15 min. after infusion	44.3	2,950	—	2.27	20	5	10
7	42	1.71	Control	43.9	2,390	490	2.20	22	5	10
			1,000 cc. in 11 min.	37.8	2,780	710	1.98	—	—	15
			30 min. after infusion	40.2	2,610	620	2.22	19	5	10
8	34	1.95	Control	52.3	2,890	620	2.01	22	7	15
			950 cc. in 6½ min.	45.2	3,280	500	2.05	35	15	23
			30 min. after infusion	44.4	3,330	—	—	32	11	20
9	25	1.93	Control	44.1	2,620	490	1.80	20	5	11
			950 cc. in 7½ min.	38.1	3,040	520	1.63	27	7	18
10	44	1.76	Control	45.7	3,200	—	2.35	17	6	11
			950 cc. in 8½ min.	44.8	3,260	—	2.42	24	10	16
			30 min. after infusion	43.1	3,390	—	—	23	10	15
11	21	1.71	Control	46.9	2,870	660	—	23	7	15
			900 cc. in 9 min.	42.4	3,170	620	—	35	15	24
			40 min. after infusion	44.9	2,990	560	—	25	7	15
12	49	1.83	Control	47.9	—	560	—	27	7	14
			900 cc. in 8½ min.	42.0	—	690	—	33	12	20
Averages	36	1.74	Control	43.0	2,700	560	2.01	21	5	12
			End of infusion	37.6	3,080 } p .001	630 } .2>p>.1	1.91	30	12	20 } p .001
			After infusion	40.0	3,030	590	2.03	23	7	13

TABLE I—*Continued*

Patient	Age	Surface area	Remarks	Pulmonary capillary pressure	Mean circulation time	Pulse rate	Oxygen consumption	Arteriovenous oxygen difference	Cardiac index (Fick)	Cardiac index (Dye)
	(*years*)	(*sq. m.*)		(*mm. Hg*)	(*sec.*)	(*beats/min.*)	(*cc./sq. m./min.*)	(*cc./100 cc.*)	(*l./sq. m./min.*)	(*l./sq. m./min.*)
1	39	1.82	Control	—	15.3	60	178	4.3	4.2	2.6
			1,000 cc. in 9½ min.	—	13.7	60	186	3.8	4.9	2.9
			38 min. after infusion	—	—	60	187	4.2	4.5	—
2	64	1.60	Control	—	16.5	75	175	3.4	5.2	—
			1,000 cc. in 11 min.	—	14.6	75	176	3.5	5.0	2.7
			60 min. after infusion	—	—	75	149	4.5	3.3	—
3	31	1.68	Control	—	9.9	65	142	4.1	3.6	3.0
			1,000 cc. in 10 min.	—	7.5	70	146	2.5	5.9	4.6
4	27	1.65	Control	—	8.9	100	164	3.3	5.0	2.8
			1,000 cc. in 11 min.	—	7.7	100	147	2.9	5.2	3.5
			35 min. after infusion	—	—	90	129	3.1	4.1	—
5	36	1.63	Control	—	11.4	95	150	4.3	3.5	3.5
			1,000 cc. in 11 min.	—	12.1	85	172	3.2	5.3	4.0
6	17	1.65	Control	5	11.0	90	161	4.2	3.8	3.2
			1,000 cc. in 13 min.	14	11.3	80	169	3.6	4.7	4.1
			15 min. after infusion	5	—	70	168	4.0	4.7	—
7	42	1.71	Control	5	14.4	70	110	4.5	2.5	2.0
			1,000 cc. in 11 min.	10	11.9	75	121	4.1	2.9	3.6
			30 min. after infusion	—	14.5	75	132	4.0	3.3	2.6
8	34	1.95	Control	6	13.3	75	142	5.6	2.5	2.8
			950 cc. in 6½ min.	—	11.4	70	149	4.8	3.1	2.6
			30 min. after infusion	—	—	65	150	4.6	3.3	—
9	25	1.93	Control	—	10.8	75	130	3.5	3.7	2.7
			950 cc. in 7½ min.	—	10.5	80	169	3.9	4.3	3.0
10	44	1.76	Control	—	—	75	166	5.9	2.7	—
			950 cc. in 8½ min.	—	11.1	60	167	5.5	3.0	—
			30 min. after infusion	—	—	65	162	5.9	2.8	—
11	21	1.71	Control	6	13.7	65	130	4.3	3.0	2.9
			900 cc. in 9 min.	15	7.8	65	161	3.9	4.2	4.8
			40 min. after infusion	8	10.6	65	134	4.6	2.9	3.2
12	49	1.83	Control	4	11.4	80	151	4.4	3.4	3.0
			900 cc. in 8½ min.	12	12.9	80	154	4.3	3.6	3.2
Averages	36	1.74	Control	—	12.4 } .2>p>.1	80	150 } .2>p>.1	4.3 } .7>p>.6	3.6 } .05>p>.02	2.9 } .02>p>.01
			End of infusion	—	11.0 }	80	160 }	3.8 }	4.3 }	3.6 }
			After infusion	—		70	151	4.4	4.4	—

sented an increase in plasma volume of approximately 385 cc. per square meter in an average subject.

In 10 cases the pulmonary blood volume showed an average post-infusion increase of 12 per cent with variations from − 6 to + 45 per cent of the control value. Both before and after the infusion the pulmonary blood volume amounted to about 20 per cent of the general blood volume. Individually, measured increases in the pulmonary and the general blood volumes were not always proportional. Five of eight subjects showed a decrease in the vital capacity at the end of the infusion.

The response of the cardiac output to the infusion was variable. The minute oxygen consumption rose slightly after the infusion. The arteriovenous oxygen difference and the central circulation time in general were somewhat reduced by the infusion. In six cases there was a significant increase in the cardiac output after the infusion while six remained within the limits of experimental error. In six out of eight cases, the third cardiac output was the same as or below the control level.

The pulse rate remained remarkably constant during the control and rapid infusion periods. In four patients repeated direct arterial pressure measurements failed to demonstrate a significant change following the saline infusion. Distention of the peripheral veins was regularly observed. Flushing of the skin was not seen. None of the subjects complained of dyspnea.

DISCUSSION

In every instance the pulmonary arterial and pulmonary capillary pressures were promptly and strikingly increased by the infusion of physiologic saline solution. A similar response has been observed following the transfusion of whole blood and the infusion of isotonic glucose and dextran solutions (8). Conversely, observations in this laboratory have demonstrated that a reduction of the effective circulating blood volume, caused by the application of venous-occluding tourniquets about the thighs, produces a lowering of the pulmonary arterial pressure (9). These data suggest that an abrupt, if not large, alteration in blood volume is the common denominator producing the observed pulmonary pressure changes.

On theoretical grounds the rise in pulmonary arterial pressure following an infusion might be the result of an increased cardiac output and therefore an increased pulmonary blood flow; of increased pulmonary resistance due to vasoconstriction within the lungs; or of an increased volume of blood contained within the pulmonary vascular bed.

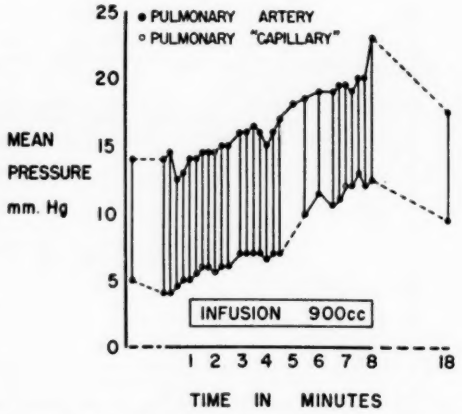

FIG. 1. SIMULTANEOUS MEASUREMENT OF PULMONARY ARTERIAL AND CAPILLARY PRESSURES DURING A RAPID SALINE INFUSION

There was no significant change in cardiac output.

The fact that the rise in pulmonary arterial pressure was the result of a rapid infusion suggested that the pressure elevation might be the result of increased flow through the pulmonary circuit. Although an increased cardiac output, and therefore an increased pulmonary flow, did occur in six instances, in the other six there was only an insignificant change in the cardiac output, yet the hypertension also resulted. In addition, there was no correlation between the magnitude of the increase in the pulmonary arterial pressure and the changes in cardiac output which did occur. This finding is not entirely surprising because previous observations on normal subjects during light exercise (10) and on patients during temporary occlusion of arteriovenous fistulas (11) have demonstrated that a considerable change in the cardiac output may occur without any alteration in the pulmonary arterial pressure.

As in the case of the pulmonary arterial pressure, there was an increase in the so-called pulmonary capillary pressure in all the patients observed. Although continuous and simultaneous

pressure readings of this function and of the pulmonary arterial pressure were not usually technically possible, intermittent readings gave evidence that there was little or no change in the pressure relationships between the two areas. In one experiment two catheters were introduced, one measuring the pulmonary arterial and the other pulmonary capillary pressure. It was demonstrated that a saline infusion raised both pressures, the gradient between the two remaining constant (Figure 1). The evidence of an elevated pulmonary capillary pressure indicates that quite probably the pulmonary venous pressure is also increased. Indeed, it may be that the pressure measured by this technique actually represents the left atrial pressure rather than the pulmonary capillary pressure (12), for the contours often show typical a-c-v waves. Further support to the concept that intravenous infusions raise the venous as well as the arterial pressure in the pulmonary circuit has been obtained from animal experiments in which both pressures were simultaneously measured with catheters in the pulmonary artery and the pulmonary vein (13). With large infusions it was found that both pressures rise, and that the venous pressure eventually approaches the elevated pulmonary arterial pressure. These data, therefore, indicate that the intravenous infusions in our patients produced a rise in the pulmonary arterial, capillary and venous pressures without gross alteration in the pressure relationships between these three components of the pulmonary circuit.

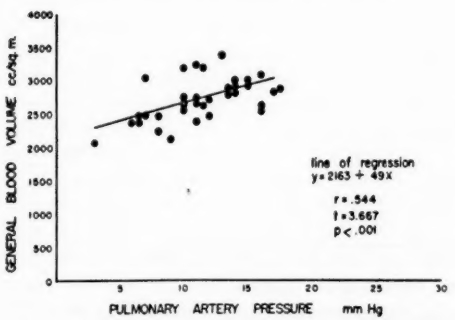

FIG. 2. THE RELATIONSHIP BETWEEN THE GENERAL BLOOD VOLUME AND THE PULMONARY ARTERIAL PRESSURE IN SUBJECTS WITH NORMAL CARDIOPULMONARY FUNCTION UNDER RELATIVELY BASAL CONDITIONS

TABLE II

Observations on patients with normal cardiopulmonary functions under relatively basal conditions

Patient	Age	Surface area	Hematocrit reading	Blood volume	Pulmonary arterial pressure	Cardiac index (Fick)
	(*years*)	(*sq. m.*)		(*cc./sq. m.*)	(*mm. Hg*)	(*l./min./sq. m.*)
1	39	1.55	38.4	2,710	11.0	4.5
2	33	1.59	41.6	3,080	16.0	6.2
3	59	1.62	28.1	2,360	6.0	5.3
4	43	1.48	34.0	2,700	12.0	2.4
5	35	1.76	49.0	3,030	7.0	3.3
6*	17	1.65	47.5	2,750	10.0	3.8
7	53	1.94	44.0	3,230	11.0	3.3
8	24	1.66	41.7	2,980	14.0	3.6
9	20	2.04	38.1	2,850	14.0	4.7
10	18	1.82	48.0	2,630	10.0	3.4
11	24	1.96	31.6	2,400	6.5	3.0
12	56	1.68	39.0	2,120	9.0	1.7
13*	25	1.93	44.1	2,620	11.5	3.7
14*	34	1.95	52.3	2,830	17.0	2.8
15*	42	1.71	43.9	2,390	11.0	2.5
16	42	1.96	44.9	2,860	17.5	4.6
17*	64	1.60	32.3	2,980	15.0	5.2
18*	31	1.68	37.7	2,450	12.0	3.6
19	21	1.88	43.8	2,970	14.0	3.8
20*	27	1.65	40.0	2,590	10.0	5.0
21	39	1.85	36.5	2,050	3.0	2.4
22	63	1.61	47.5	2,940	15.0	4.0
23	49	1.91	36.8	2,610	16.0	3.6
24*	39	1.82	42.4	2,430	6.5	4.2
25	59	1.77	43.9	3,380	13.0	
26	31	1.30	27.0	2,710	11.0	5.5
27	34	1.87	32.9	2,250	8.0	4.8
28	44	1.40	34.0	2,560	16.0	2.7
29*	44	1.76	45.7	3,200	11.5	2.7
30	34	1.51	38.5	2,470	8.0	5.1
31*	21	1.71	46.9	2,870	13.5	3.0
32*	36	1.63	34.7	2,470	7.0	3.5
33	47	1.82	39.2	3,180	10.0	3.6
34	28	1.66	48.5	2,820	13.5	4.3
Mean	37.5	1.73	40.4	2,720	11.4	3.8
S.D.				±316	±3.49	±1.03

* Denotes patients receiving saline infusions and included in Table I.

The maintenance of essentially normal pressure gradients throughout the pulmonary circuit in subjects receiving rapid infusions is considered as evidence against the occurrence of pulmonary arteriolar constriction as the cause of the pressure changes in these subjects. Had arteriolar constriction been the cause of the increased pulmonary arterial pressure, little or no increase in the pulmonary capillary pressure would have been observed. Further evidence against vasoconstriction was obtained by examination of the contour of the pulmonary arterial pressure tracings which revealed no indication of decreased "run-off" during diastole.

If increased flow and increased resistance due

to arteriolar constriction may be eliminated as the fundamental mechanisms producing the pulmonary hypertension, it becomes necessary to explore the possibility of an increased volume of blood in the lungs.

First, the possible relationship of the pulmonary arterial pressure to the volume of blood in the pulmonary circuit under relatively basal conditions was investigated. A study of the so-called "pulmonary blood volume" failed to show good correlation with the pulmonary pressures. On the other hand, the relationship of the pulmonary arterial pressure to the general blood volume revealed a striking correlation. This is demonstrated in Figure 2, where in addition to the observations made in this study, there are included for further statistical validation 22 points derived from studies by the same methods on additional patients with normal cardiopulmonary function made under relatively basal conditions (Table II). A linear relationship of good statistical significance is demonstrated. If the results obtained on the effect of saline infusions are plotted in a similar way, they will be seen to follow closely the line of regression derived from the observations on normal subjects under basal conditions (Figure 3). Although the line of regression is represented as straight, it is very possible that in actuality it is curved and follows more nearly the pressure-volume relationship shown by elastic structures or, more particularly, by isolated segments of veins (14, 15). Apparently, however, under the conditions of our studies, the relationship between the blood volume and the pulmonary arterial pressure is nearly linear. Infusion of one liter of saline solution in our human subjects, however, gave no evidence of the disproportionately large rise in pulmonary pressures such as reported in the open-chested dog by Opdyke and associates (16).

The studies reported here indicate that the ratio between the quantities of blood contained in the pulmonary and the systemic circuits is not altered by a rapid expansion of the general blood volume. Yet, because of the pressure-volume relationship present in the pulmonary venous-left atrial segment, this small increase in blood content produced a considerable increase in pulmonary pressures. Similar evidence in the opposite direction is available from the study of blood loss in normal human subjects (8). If there were disproportionate addition to or loss from the pulmonary circulation, or if pulmonary vasomotor activity were stimulated, the relationship demonstrated in Figure 2 would not hold and the plotted

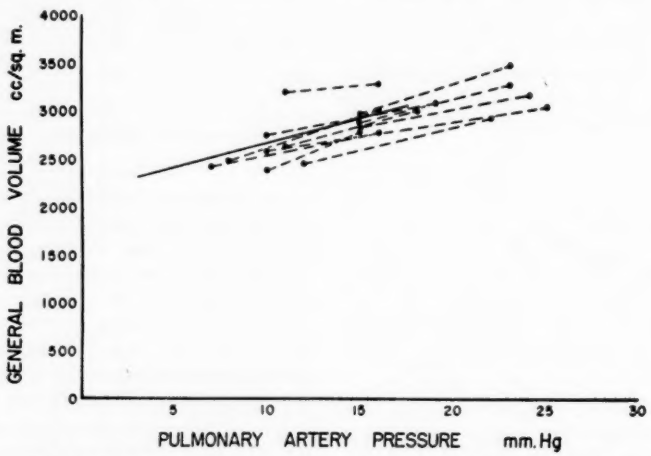

FIG. 3. THE RELATIONSHIP OF THE GENERAL BLOOD VOLUME AND THE PULMONARY ARTERIAL PRESSURE IN NORMAL SUBJECTS RECEIVING SALINE INFUSIONS

In each case the pulmonary arterial pressure rises. The line of regression is that obtained from Figure 2.

results would not fall on the line of regression. During hypoxia in normal subjects and in certain pathologic situations such a disproportion does occur (8). These pressure relationships are evidence against, but do not eliminate the unlikely possibility that our infusions produced acute cardiac failure.

The reason for the absence of a good correlation between the observed pulmonary arterial pressure and the measured pulmonary blood volume in our studies is not apparent. The mean values show the expected directional trend but the individual values appear to be somewhat erratic. It is felt probable that changes in the pulmonary volume of the order of 50–100 cc., as induced in many of these experiments, are not sufficiently beyond the range of error in the measurement itself (8).

Insofar as one can measure the filling pressure from observations made at one point, it is interesting to note that despite the evidence of an increase in the pulmonary venous, and therefore the left heart filling pressure, there was not a consistent increase in the left ventricular output. These data are compatible with the concept that alterations in the filling pressure are not a major factor in producing changes in the cardiac output in the human being (17).

It has been demonstrated previously that intravenous infusions similar to those utilized here produce an increase in the right ventricular, right atrial, central venous and peripheral venous pressures (8, 18). Although simultaneous readings of these pressures and those in the pulmonary circuit are not available, the evidence indicates that the rise produced is essentially the same throughout the venous and pulmonary circuit. The normal pressure gradients appear to be maintained thereby from peripheral vein to left atrium, but at a higher level following the infusion.

In the present study the pulse rate remained remarkably constant from the control through the infusion period. This failure to observe the Bainbridge reflex in the normal human has been observed previously in this laboratory (18).

It is difficult to assess the relationship of the pulmonary hypertension produced in these studies to that occurring in patients with cardiac disease. These experiments, as well as other recently reported observations (19, 20), demonstrate the occurrence of elevations in the pulmonary capillary pressure of such magnitude as adequately to account for the production of pulmonary edema without implication of changes in capillary permeability. Further studies are under way on the effect of manipulations similar to those described here on the pulmonary hemodynamics of patients with cardiac and pulmonary disease.

SUMMARY

In the normal human subject the pulmonary arterial and the pulmonary capillary pressure can be significantly increased by the rapid intravenous infusion of physiologic saline solution. The pulmonary and the general blood volumes increase proportionately after this maneuver. The cardiac output after a rapid infusion may or may not increase.

There is a linear correlation between the resting pulmonary arterial pressure and the general blood volume in normal subjects but this relationship is not apparent in the case of the pulmonary blood volume as measured by the dye technique. The same relationship continues after an increase in the blood volume with saline solution. It is suggested that the changes induced by a rapid infusion can be explained most adequately on the basis of an abrupt, but not disproportionate, increase in the amount of blood contained in the relatively indistensible pulmonary circuit.

ACKNOWLEDGMENT

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COMPARISON OF INTRACARDIAC AND INTRAVASCULAR TEMPERATURES WITH RECTAL TEMPERATURES IN MAN [1, 2]

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(Submitted for publication September 28, 1950; accepted, January 22, 1951)

A study of intracardiac and intravascular temperatures was undertaken with the thought that such measurements might provide further information concerning the mechanisms which normally maintain body temperature within its narrow limits and also that they might help to explain the deviations from normal temperature in certain disease states. Whereas the temperature of the peripheral tissues may vary widely, homeothermic man maintains the temperature of the deep tissues, or of a critical deep tissue, fairly constant under widely varying conditions of heat loss and heat gain (2). The identity of the critical deep tissue is not completely understood but a considerable body of evidence places it in the diencephalon and in normal man small variations in its temperature are thought to initiate compensating mechanisms which return the temperature of the body to the homeothermic norm. Generally, rectal temperature has been considered to indicate deep tissue, and critical tissue, temperatures but often rectal temperature appears to be a lagging and unsatisfactory index; witness the onset, when the body is heated, of such compensating mechanisms as peripheral vasodilatation and sweating before a rise in rectal temperature has occurred; and conversely, in subjects who are cooled, the presence of vasoconstriction and pilomotor activity before a fall in rectal temperature has taken place. It seemed desirable, therefore, to look for some other index of critical deep tissue temperature.

Considerations of the mixing of blood in the left ventricle, probably in proportion to the volume flow and temperature of the blood from all organs, suggested the temperature of left ventricular blood as representative of deep tissue temperature. The difficulties in such a measurement, together with the likelihood that no significant change in temperature occurs during the transit of blood through the aorta, lead to the substitution of the temperature of the blood high in the femoral artery for left ventricular temperature. The observation (3) that such femoral arterial temperatures consistently fell below simultaneously recorded rectal temperatures, and that rectal temperature lagged behind changes in temperature of femoral arterial blood during changing body temperature, prompted the measurement of more "central" blood temperatures.

It was the purpose of the present study to determine the intracardiac and deep intravascular temperatures in afebrile subjects and thus establish a baseline of values with which future determinations, obtained during abnormal and changing body temperatures, could be compared.

METHODS

Observations were made on 24 afebrile hospitalized subjects, usually several hours after breakfast and occasionally after an overnight fast. Most of the determinations were made at ambient temperatures between 77° F. and 84° F. and with the subjects nude, except for a loin cloth. During the few observations made at lower room temperatures the subjects were covered with one sheet.

All temperatures were measured potentiometrically with naked-junction, copper-constantan thermocouples and a Leeds-Northrup, Type K2, potentiometer. The temperatures in the chambers of the right heart and in the deep vessels leading to the right heart were measured by a thermocouple (copper wire gauge 30, constantan wire gauge 30) threaded to the tip of a standard, 8F, intracardiac catheter. This was a roving thermocouple, introduced in varying succession by the technique of cardiac catheterization (4), and under fluoroscopic control, into the right heart and connecting vessels. In determining a temperature with this thermocouple, it was not moved to a new location until three successive read-

[1] Read in part at the Thirty-third Annual Meeting of the American Physiological Society, Detroit, Michigan, April 21, 1949 (1).

[2] This study was supported by a grant from the Baruch Committee on Physical Medicine.

ings, made over a period of five to 10 minutes, checked each other.

All other thermocouples were kept stationary after they were positioned properly. Femoral arterial and femoral venous blood temperatures were obtained by indwelling, plastic-sheathed, thermocouples (copper wire gauge 42, constantan wire gauge 30) threaded through Cournand needles previously inserted into the corresponding vessels high in the groin (3, 5). The thermal junctions of these thermocouples extended in the lumen of the vessels for a distance of 5 to 6 centimeters beyond the needle tips and probably reached the junction of the femoral with the iliac vessels. Rectal temperature, 3 inches above the anal orifice, was determined by a single thermal junction (copper wire gauge 30, constantan wire gauge 30) presenting on the lateral wall of a suppository-shaped, hard-plastic applicator. Subcutaneous temperatures immediately beneath the skin, and intramuscular temperatures at a depth of 1½ inches below the skin surface, were determined in the lateral aspect of the thigh by means of gauge 22 needle thermocouples (copper wire gauge 42, constantan wire gauge 30). Because of their small masses all thermocouples had rapid response times, usually 95% of full deflection in 10 to 15 seconds. At the close of each experiment the calibration factor of each thermocouple was determined for that experiment.

RESULTS

The data for a typical observation period are plotted in Figure 1, which shows the relationships of the various intracardiac and intravascular temperatures to each other, and to rectal temperature. These relationships are shown more definitively in Table I and in Figure 2, wherein the data for all of the observations are analyzed with reference to the rectal temperature at the time. The data are consistent and indicate several thermal gradients.

The temperatures in the right heart (atrium and ventricle) and pulmonary artery (main stem and

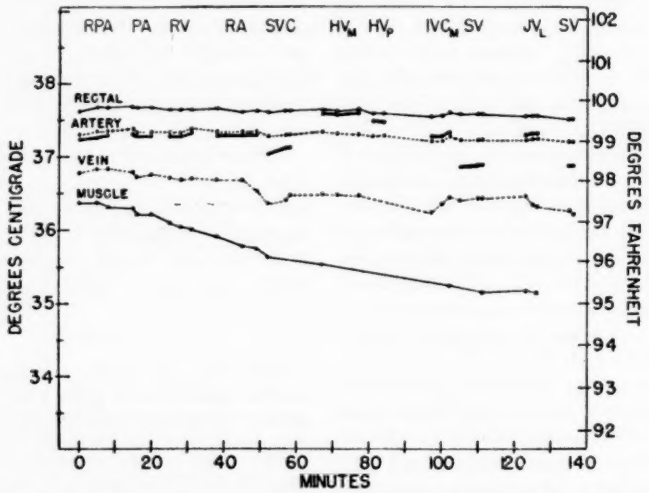

FIG. 1. OBSERVATIONS FROM A TYPICAL EXPERIMENT SHOWING THE RELATIONSHIPS OF INTRACARDIAC AND INTRAVASCULAR TEMPERATURES TO EACH OTHER AND TO RECTAL TEMPERATURES (AFEBRILE SUBJECT)

The lines from above downward indicate in turn, rectal temperature, femoral arterial temperature, femoral venous temperature and intramuscular (thigh) temperature. The heavy, disconnected, solid bars indicate the temperatures obtained with the catheter thermocouple in the locations indicated directly above by the letters at the top of the graph. In this and subsequent figures these letters indicate: RPA, right pulmonary artery; PA, main stem of pulmonary artery; RV, right ventricle; RA, right atrium; SVC, superior vena cava; SV and SC subclavian vein; J_L and JV_L, jugular vein, low; J_h and JV_h, jugular vein, high; IVC_L, inferior vena cava, low; IVC_m, inferior vena cava, mid-portion; IVC_h, inferior vena cava, high; H_p and HV_p, hepatic vein near vena cava; HV_m, hepatic vein, mid-liver; H_d and HV_d, hepatic vein deep in the liver; AV, axillary vein.

TABLE I

Analysis of the deviations of intracardiac and intravascular temperatures from rectal temperature in afebrile subjects

Location of catheter thermocouple	Number of patients	Difference between "catheter" temperature and rectal temperature*			
		Mean °C.	Standard deviation °C.	Fisher t value	Probability† P
Right Pulmonary Artery (RPA)	16	−0.27	0.17	6.24	0.01
Pulmonary Artery Main Stem (PA)	13	−0.26	0.15	6.19	0.01
Right Ventricle (RV)	17	−0.23	0.14	6.57	0.01
Right Atrium (RA)	24	−0.26	0.13	9.96	0.01
Superior Vena Cava (SVC)	19	−0.35	0.16	9.01	0.01
Subclavian Vein (SV)	21	−0.58	0.22	11.55	0.01
Jugular Vein, low (J_L)	15	−0.22	0.15	5.51	0.01
Jugular Vein, high (J_h)	4	−0.01	0.04	0.22	0.80
Inferior Vena Cava, low (IVC_L)	7	−0.26	0.22	2.84	0.02
Inferior Vena Cava, high (IVC_h)	10	−0.22	0.18	3.63	0.01
Hepatic Vein, near Vena Cava (H_p)	6	−0.09	0.10	1.88	0.10
Hepatic Vein, deep in liver (H_d)	6	−0.03	0.04	1.84	0.10
Femoral Artery (FA)	22	−0.22	0.15	6.66	0.01

* Minus indicates catheter temperature less than rectal temperature.
† Due to the small size of the samples, P values are not expressed smaller than 0.01 even though the P often calculated to a considerably smaller value.

right branch) were practically identical, essentially equal to femoral arterial temperature, and less than rectal temperature by an average of 0.25° C. (0.45° F.). Analysis shows that this difference between right heart and rectal temperature, though small and with a sizable standard deviation, is nonetheless statistically significant (Table I) because the intracardiac temperatures so consistently fell below rectal temperature (Figure 2).

Progressively lower temperatures were recorded as the thermocouple was moved from the right atrium into the superior vena cava and into the subclavian vein, where the temperature averaged 0.6° C. (1.1° F.) below rectal temperature (Figure 2). Statistically, the temperatures in both of these vessels are significantly below rectal temperature (Table I). In the inferior vena cava there were inconstant variations from right atrial temperature but on the average the temperature at all levels was approximately equal to that in the right atrium (Table I, Figure 2).

Two sites contributing blood of higher temperature to the circulation were noted, the brain and the liver. As the tip of the thermocouple passed from the subclavian vein into the common jugular vein, even for a distance of only a few centimeters, the blood temperature rose sharply from a relatively low value to a value equal to right heart temperature (JV_L, Figure 1). High in the jugular vein, approximately in the jugular bulb, the temperature was still higher, well above right heart temperature and equal to rectal temperature (Table I, Figure 2). Similarly the temperature of hepatic vein blood exceeded that of the inferior vena cava and equalled rectal temperature (Table I, Figure 2). The observations on hepatic and jugular blood temperatures were few in number but analysis suggests that these temperatures are probably the same as rectal temperature (Table I).

In brief, rectal temperature was slightly higher than the intracardiac and deep intravascular temperatures here measured, except that high in the jugular vein and deep in the hepatic vein the temperatures equalled rectal temperature.

In 20 afebrile subjects the temperature in the femoral artery was found to be similar to that in the right heart (Table II, Figure 3). Moreover, femoral arterial temperature was essentially the same as the temperature in most of the deep vessels. However, in the hepatic vein, and high in the jugular vein, the temperature was higher than femoral artery temperature and in the superior vena cava and subclavian vein the temperature was lower (Table II, Figure 3).

The differences between rectal and right heart temperatures were so small in afebrile subjects that for practical purposes the two temperatures may be considered to be the same. The few preliminary observations thus far made indicate that such a similarity may not be the case when body

TABLE II

Analysis of the deviations of intravascular and intracardiac temperatures from femoral arterial temperature in afebrile subjects

Location of catheter thermocouple	Number of patients	Difference between "catheter" temperature and femoral artery temperature*			
		Mean °C.	Standard deviation °C.	Fisher t value	Probability† P
Pulmonary Artery Main Stem (PA)	16	−0.03	0.07	1.36	0.2
Right Ventricle (RV)	17	−0.01	0.06	0.46	0.6
Right Atrium (RA)	20	−0.03	0.07	1.84	0.1
Superior Vena Cava (SVC)	17	−0.11	0.08	5.83	0.01
Subclavian Vein (SV)	18	−0.36	0.16	9.15	0.01
Jugular Vein, low (J_L)	11	+0.02	0.12	0.62	0.5
Jugular Vein, high (J_h)	4	+0.15	0.21	1.26	0.3
Inferior Vena Cava, low (IVC_L)	4	−0.08	0.17	0.85	0.5
Inferior Vena Cava, high (IVC_h)	9	+0.03	0.13	0.58	0.6
Hepatic Vein, proximal (H_p)	3	+0.11	0.10	1.56	0.3
Hepatic Vein, deep (H_d)	4	+0.26	0.05	9.37	0.01

* Plus indicates catheter temperature higher than rectal temperature; minus, catheter temperature less than rectal temperature.

† See footnote, Table I.

temperature departs from the homeothermic norm. The data plotted in Figure 4 were obtained in a febrile patient with slowly resolving pneumonia and show a very sizable difference (0.8° C., 1.4° F.) between rectal and right heart temperatures. Furthermore, in five febrile patients with rectal temperatures ranging from 38.2° C. to 40.0° C. (100.7° F. to 104.0° F.) the differences between right heart and rectal temperatures were 0.2°, 0.4°, 0.6°, 0.7° and 0.8° C. Right heart temperature

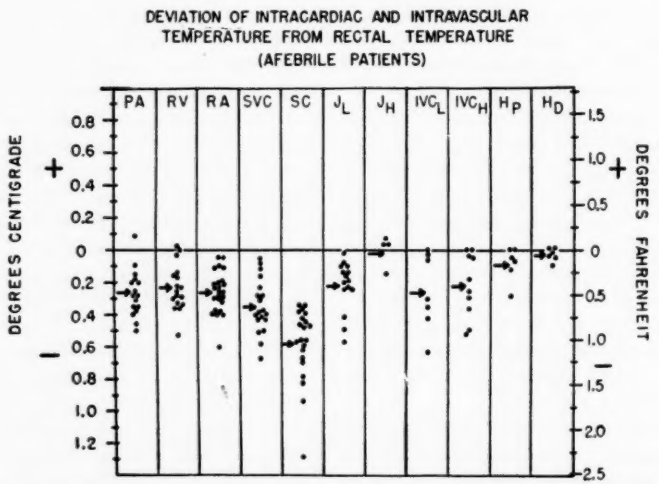

FIG. 2. INTRACARDIAC AND INTRAVASCULAR TEMPERATURES PLOTTED WITH REFERENCE TO THE SIMULTANEOUSLY OBTAINED RECTAL TEMPERATURE AS A BASE (AFEBRILE SUBJECTS)

Each dot represents a comparison in one subject between rectal temperature and the intravascular temperature indicated by the letters at the head of the respective columns. Each heavy horizontal arrow represents the average of the corresponding group of dots.

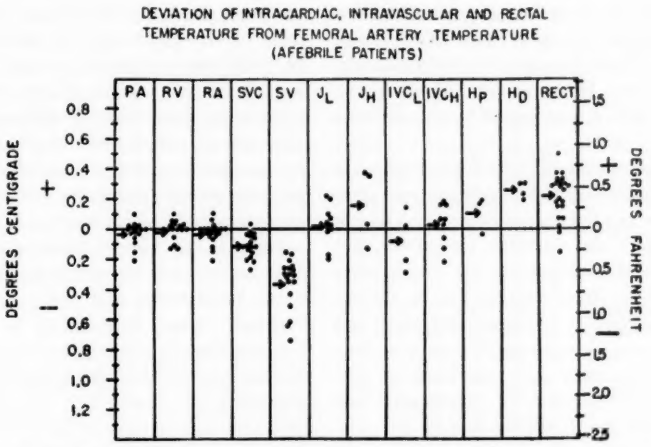

FIG. 3. INTRACARDIAC, INTRAVASCULAR AND RECTAL TEMPERATURES PLOTTED WITH REFERENCE TO THE SIMULTANEOUSLY OBTAINED FEMORAL ARTERIAL TEMPERATURE AS A BASE (AFEBRILE SUBJECTS)

Details as in Figure 2.

was always less than rectal temperature and the difference was greater in the patients with the higher fevers. In all five patients the relationships of the various intravascular temperatures to each other and to rectal temperature were the same as in afebrile subjects, and again, femoral arterial temperature was the same as right heart temperature.

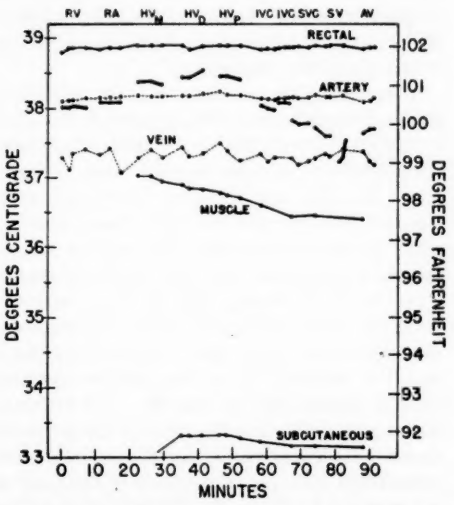

FIG. 4. RELATIONSHIP OF INTRACARDIAC AND INTRAVASCULAR TEMPERATURES TO EACH OTHER AND TO RECTAL TEMPERATURE IN A FEBRILE PATIENT WITH SLOWLY RESOLVING PNEUMONIA

Details as in Figure 1.

DISCUSSION

The data indicate a small, but consistent, gradient of increasing temperature in the larger veins as they approach the heart. The gradient is steeper in the thorax than in the abdomen where, at times, it is not demonstrable. Sharp rises in temperature above these gradients occur within veins which drain organs considered to have increased metabolic heat production. The liver and brain are shown to be such organs.

It was not anticipated that femoral arterial temperature would be the same as right heart temperature. When previous observations (3) showed that femoral arterial temperature fell below rectal temperature, the supposition was held that right heart temperature probably equalled, or exceeded, rectal temperature and that the lower femoral arterial temperature was due to the cooling of blood within the lungs (assuming again that blood does not change in temperature during transit through the aorta). Since right heart and femoral arterial temperatures are essentially the

same, it follows that heat loss from the lungs is *relatively* so small with respect to the large volume of blood which flows through the lungs that the temperature of the blood is not altered by an amount which can be determined by the methods here used.

Since the temperature of right heart blood was consistently less than rectal temperature, and since this difference was small, a systematic variation in the thermocouples was carefully looked for, but excluded. The consistency of the temperature pattern and the statistical analysis lead us to conclude that the difference between right heart and rectal temperature, though small, is a real one. Since the measurements were not made at controlled ambient conditions the significance and constancy of this small difference may be questioned, for it is well known that the temperatures of various portions of the body may vary considerably at different environmental temperatures and humidities. In this regard attention is called to the relatively narrow range (7° F.) of the ambient temperatures, all of which were in a zone comfortable for the nude subject. Furthermore, regardless of the variations in temperature and humidity within this range the relationship of the rectal and intracardiac temperatures was always the same, with respect to both the direction and the degree of difference.

It seems reasonable that there should be a difference between rectal and right heart temperatures since one gives the temperature of a single organ whereas the other is influenced by thermal contributions from all organs. It seems reasonable, too, that femoral arterial temperature should be similar to intracardiac temperature, since both determinations are measurements of mixed blood temperature. Furthermore, differences between intracardiac, intravascular and rectal temperatures of the magnitude and direction here reported have been demonstrated in animals; originally by Claude Bernard (6), who introduced thermometers into the heart and various vessels, and more recently by Horvath, Rubin and Foltz (7) who used intravascular thermocouples introduced under fluoroscopic control.

Although the differences between right heart temperature and rectal temperature were not sufficiently large to be of practical importance in afebrile subjects, these differences assume significance in two respects; in febrile patients and in considerations of the physiology of thermal homeostasis. In preliminary observations on febrile patients, right heart temperature at times fell so far below rectal temperature that the differences between the two cannot be dismissed. Further studies of thermal gradients within the vascular tree during fever may serve to clarify the factors responsible for maintaining the febrile state.

In regard to thermal homeostasis, attention has focussed on two temperatures; the average deep tissue temperature and the temperature of that "critical" tissue, believed to be located in the hypothalamus, which triggers the thermoregulating mechanisms which return body temperature to the homeothermic norm. Present concepts generally hold that both of these temperatures are measured by rectal temperature. This supposition appears open to question. The observations here reported confirm that variations in temperature exist even deep within the body core and this precludes the temperature of a single organ as the measure of *average* deep tissue temperature. An integrating measurement or a calculated average value seems more likely to indicate this temperature. Furthermore, evidence is not at hand to prove that the temperature of the rectum is the same as that of the critical tissue, particularly during conditions of changing body temperature.

Intracardiac temperature, and its equivalent femoral arterial temperature, appears to represent a "vascular" integration of the various individual temperatures to give an average blood temperature. This temperature seems to approach *average* deep tissue temperature more closely than any other single measurement but, because of the thermal contributions from the peripheral tissues, it may not be the same as average deep tissue temperature. Since the critical tissue temperature is the temperature of an organ, it is not likely that it would be indicated by an integrated temperature such as intracardiac temperature. Nevertheless, intracardiac and femoral arterial temperatures should have considerable significance in thermal homeostasis since the temperature of the blood as it enters, and affects, the critical tissue in the hypothalamus is more apt to be indicated by these temperatures than by any other measurement now made. Only the temperature of the blood in the afferent artery to the critical tissue would have

greater significance and this is a measurement not now available.

The above considerations, based on the data presented, are suggested for the afebrile subject in thermal equilibrium. It remains for future study to determine whether the relationships between intracardiac, intravascular and rectal temperatures here reported, and the considerations drawn therefrom, also hold under the more critical conditions of changing body temperature, when thermoregulatory mechanisms are actually called into action.

SUMMARY

1. Measurements of the temperature in the right heart, in the deep vessels leading to the right heart, in the femoral artery and in the rectum of 24 afebrile subjects showed:

(*a*) there is a gradient of increasing temperature in the larger veins as they approach the heart,

(*b*) temperatures within the right heart and pulmonary artery are equal, and the same as the temperature high in the femoral artery,

(*c*) rectal temperature exceeds intracardiac and deep intravascular temperatures by a small but significant amount,

(*d*) temperatures in the veins draining the liver and brain are higher than the temperatures in the veins into which they drain, and equal rectal temperature.

2. The differences between rectal and intracardiac temperatures are not of such magnitude as to be of clinical importance in afebrile subjects.

3. Preliminary observations suggest that during fever rectal temperature may exceed intracardiac temperature by a significant amount, up to 0.8° C. in one patient.

4. The data suggest a review of those concepts of thermal homeostasis which hold that rectal temperature represents either the average deep tissue temperature, or the temperature of the critical tissue which initiates thermoregulating mechanisms.

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THE IMMUNOPHYSIOLOGY OF SERUM SICKNESS. ALTERATIONS IN THE BLOOD VOLUME AND THIOCYANATE SPACE IN RELATION TO THE DEVELOPMENT OF HUMORAL ANTIBODIES IN THE RABBIT[1]

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The development of edema during the course of acute infectious diseases has been observed in patients for many years. The retention of water and salt during the course of untreated pneumococcal pneumonia was described early in the century (1). The loss of chlorides from the blood and their subsequent excretion in the urine during recovery was shown to be associated with retention by the body of sodium and calcium while potassium and magnesium were excreted normally or in excess.

After antipneumococcal horse serum became available for treatment of pneumonia, serum sickness developed frequently in patients during convalescence. The clinical picture of serum disease with edema and fever was shown to be accompanied by the retention of chlorides and water in the body (2). Further investigation of serum disease in human beings revealed that the clinical symptoms are associated with the appearance in the blood of precipitins against the species of animal in which the therapeutic serum was prepared (3). The appearance of precipitins precedes recovery from the disease by a short interval and is coincident with disappearance of the antigen. These studies indicated that antigen-antibody reactions were of clinical significance in the production of disease in human beings, and that the mechanism was in some way related to that of experimental anaphylaxis in animals.

During the course of studies on patients with Rocky Mountain spotted fever, it was observed that alterations in the distribution of body fluids with the development of marked clinical edema reached a maximum just before clinical recovery begins. The alterations in distribution of fluid appear to predispose to peripheral circulatory collapse. In such crises the blood chlorides, blood volume and extra-vascular thiocyanate space are altered transiently but return to normal with recovery (4). Observations in one patient who had been given hyperimmune rabbit antiserum for Rocky Mountain spotted fever showed that the alterations in the blood volume and thiocyanate space, which occurred during classic serum sickness following recovery from the initial disease, resembled strikingly those seen in the same patient during the acute phase of the rickettsial infection. These observations suggested the possibility that an immune reaction may be directly or indirectly responsible for the changes in the permeability of membranes which permit alterations in fluid distribution in rickettsial spotted fever.

The present experiments were undertaken to determine if changes in the blood volume and thiocyanate space occur in an antigen-antibody reaction induced *without infection* and to relate the physiologic alterations to the development of immunity. The anatomic changes occurring with serum sickness have been well studied, but the physiologic alterations induced by the disease are very poorly understood (5). Accordingly, rabbits were injected with human plasma and the changes in distribution of body fluids were followed serially.

MATERIAL

Domestic rabbits of mixed breeds, weighing 2 to 4 kilograms each, were placed in individual cages and fed a stock diet, supplemented once weekly by fresh green vegetables. Water was given without restriction.

The same sample of pooled human plasma was used as the sensitizing antigen and as antigen for the precipitin tests in each individual experiment.

[1] This study was supported (in part) by research grants from the John and Mary R. Markle Foundation, the National Heart Institute of the National Institutes of Health, Public Health Service, and the Atomic Energy Commission.

METHODS

Chemical techniques: The *plasma volume* was determined by the T-1824 (Evans Blue) dye method. Syringes holding 2 ml. were calibrated to deliver 0.5 ml. of a solution containing 3 mg. of the dye per milliliter. The *thiocyanate space* was determined simultaneously. Syringes holding 10 ml. were calibrated, and 2.0 ml. of a solution containing 50 mg. of sodium thiocyanate per milliliter were injected.

The concentrations of the dye in rabbit serum were determined by the Evelyn photoelectric microcolorimeter and the thiocyanate concentrations with the Evelyn photoelectric macrocolorimeter (6). *Hematocrit* readings were made on oxalated blood placed in Wintrobe tubes, centrifuged at 3,000 revolutions per minute for 30 minutes.

The *total blood volume* was calculated from the plasma volume and the hematocrit.

Immunologic techniques: The humoral antibody titer was followed by ring *precipitin tests.* The human plasma was diluted 1:100 in water and used as antigen to overlay rabbit serum serially diluted in 2 × 20 mm. tubes to a maximum of 1:32. In the definitive experiment human plasma was diluted in physiologic saline, but the rabbit serum was undiluted. In the preliminary experiments dilution of human plasma (antigen) as high as 1:100,000 gave positive tests with undiluted rabbit serum (antibody) as definite as those obtained with 1:100 dilution of antigen and 1:32 dilution of antibody.

The dye was diluted as much as 1:50,000 for precipitin tests in group 1; no precipitins were encountered, so the test was not performed in later groups.

Skin tests to the dye were never positive in group 1; so they were not performed in later groups. No skin tests were done with plasma in any group.

Experimental procedure: In the morning before feeding the animals were weighed and strapped on their backs to wooden boards. In groups 1 and 2 blood was withdrawn without anesthesia in an oiled syringe by cardiac puncture.

Comparison of data indicated that reliable results could be obtained without deaths from hemopericardium by nicking an ear vein with a knife and collecting the blood in dry test tubes. Accordingly in group 3, 2 ml. were collected and allowed to clot; 1 ml. was then collected in another dry tube containing 6 mg. of ammonium oxalate and 4 mg. of potassium oxalate. The dye was injected into a vein in the opposite ear, the syringe changed, and the sodium thiocyanate injected through the same needle within one minute. Ten minutes following completion of the injection, 2 ml. of blood were collected from the first ear.

Preliminary experiments: No data were available on the reliability of the technical methods in small animals, on the effect of repeated bleedings at short intervals, or on the normal values for plasma volume and thiocyanate space in rabbits. It was not known whether rabbits might become sensitized after repeated injections of the dye and hence develop a picture analogous to serum sickness.

Accordingly, six animals (group 1) were used as a control group on the technical procedures. Determinations of the blood volume, thiocyanate space and precipitin titers, as well as a skin test, were done simultaneously every second or third day. The animals received no plasma. They were kept under observation for a minimum of 30 and a maximum of 46 days. A minimum of nine and a maximum of 16 determinations were done on each individual animal.

A second experiment was performed in order to study the variability of the physiologic changes in individual animals ill with serum sickness. It was also necessary to determine the optimum route of sensitization, the required dose of plasma and the time relationship of any observed alterations to the development of humoral immunity.

Accordingly, 11 animals (group 2) were injected with plasma. The two groups were treated in the same fashion except for the sensitizing injections of plasma. After a baseline for the individual animal had been established, the rabbits in group 2 received injections of plasma ranging from 2.2 to 5.0 ml. per kilogram of body weight. Eight animals received the sensitizing dose intravenously and three were injected subcutaneously in the intrascapular region. Three of the former animals received a second injection intravenously on the day following the initial injection.

During the clinical peak of the disease, determinations of the blood volume and thiocyanate space were done on two successive days. All animals in group 2 were followed at least 24 days following the injection of plasma, unless accidental death from hemopericardium supervened. A minimum of seven determinations were done on each animal in group 2 after the baseline was established.

In order to interpret the data critically a satisfactory statistical method of analysis had to be devised. It was necessary to determine whether groups could be compared or whether each individual animal must serve as its own control. The data from groups 1 and 2 were analyzed by several techniques and the results obtained were submitted to an impartial arbiter for criticism.[2] A satisfactory statistical method was devised and the definitive experiment designed to improve the statistical significance of the data obtained in the preliminary experiments.

Definitive experiment: Twelve rabbits (group 3) of more uniform size, weighing approximately 2 kilograms each, were injected with human plasma. The chemical and immunologic techniques and experimental procedure as described above were followed. At least two baseline determinations were made every second day on each animal. After the second determination, each animal was injected intravenously with a single dose of 3 ml. of plasma per kilogram of weight. Precipitin tests were performed daily on each rabbit, but no further fluid vol-

[2] We are deeply indebted to the Institute of Statistics of the University of North Carolina, Raleigh, N. C., for critical review of the data and for suggesting the method subsequently used.

ume determinations were done until the day that a positive precipitin reaction was first obtained. The measurements were repeated on that day, on two consecutive days thereafter, and at varying intervals subsequently until the chemical values had returned approximately to the baseline.

Statistical analysis: The total blood volume, plasma volume, hematocrit and thiocyanate space were recorded in absolute values. The mean of two successive baseline determinations on each animal was compared with the mean of the two successive determinations made on the day the precipitins became positive and on the following day. The mean differences between the baseline values and those obtained after the precipitin test became positive were calculated. The significance of the mean difference was tested by the following formulae in which $\bar{d}$ = mean difference, $s_{\bar{d}}$ = standard error of the mean difference, S = summation of, n = number of animals:

$$t = \frac{\bar{d}}{s_{\bar{d}}} \qquad s_{\bar{d}} = \sqrt{\frac{Sd^2 - \frac{(Sd)^2}{n}}{n(n-1)}}.$$

The calculated value for t was compared with the value at $n-1$ degrees of freedom as obtained from standard tables.

The data as obtained in absolute values were then calculated in terms of milliliters per kilogram of body weight on the day the measurement was made. The statistical significance was determined by the same formulae.

In view of the fact that the experiment was planned to evaluate changes in a febrile illness occurring in a growing animal the difference in the amount of food and water ingested by the animals unavoidably introduced variables which were difficult to control. Accordingly, the data were re-calculated in terms of milliliters per kilogram of *initial* body weight (the day of the first baseline determination) in order to avoid consideration of differences in food and water intake and in order to permit ready comparison with the data in group 1. The statistical significance was then determined by the same method.

It should be noted that in the definitive experiment no attempt was made statistically to compare the experimental group as a whole with the control group. Each animal served as its own control.

RESULTS

Control on methods: None of the animals in group 1 showed any symptoms or signs suggesting serum sickness. No reaction was noted in any rabbit to the repeated intravenous injection of dye and no evidence of sensitization was detected by skin or precipitin tests. The fatal dose of T-1824 dye was found to be 3 mg. per kilogram of body weight given intravenously; death occurred within 15 minutes.

Moderate variations in *plasma volume, thiocyanate space* and *total blood volume* per kilogram of body weight and in the *hematocrit* were noted among the different animals. In serial determinations on a single rabbit, however, the results were much more uniform. When the data obtained near the end of the experiment were examined, a slight decrease in the thiocyanate space in relation to

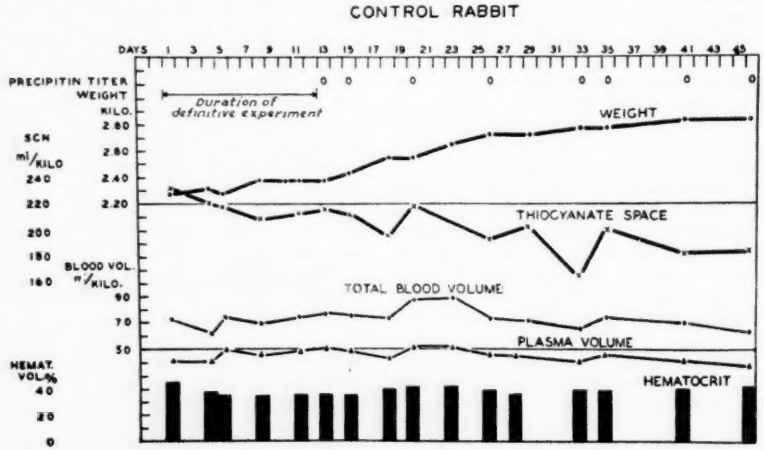

FIG. 1. FLUID SPACES IN A CONTROL RABBIT (GROUP 1) NOT INJECTED WITH PLASMA

All of the serial determinations are plotted in this graph; only four values which fall within the indicated duration of the definitive experiment are used in the calculations in Tables I and II.

TABLE I

Control of technical methods (group 1)

Rabbit number	Baseline					Time of expected response				
	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit
	Kilo.	*ml./Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*Vol. %*	*Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*Vol. %*
1	2.30	219.6	44.2	68.8	36.0	2.39	216.1	49.4	75.5	34.5
2	3.04	197.8	47.0	68.8	31.3	3.04	197.0	55.8	87.2	35.7
3	2.53	210.3	51.3	73.5	30.0	2.78	193.8	49.8	72.8	31.5
4	3.27	177.5	43.0	63.5	33.3	3.24	162.5	40.0	62.1	35.5
5	2.76	207.7	49.7	77.3	35.8	2.93	198.1	47.9	73.3	34.5
6	2.73	155.3	42.7	67.0	36.3	2.92	162.2	45.0	62.7	28.0
Mean for group	2.77	194.7	46.3	69.8	33.8	2.88	188.3	48.0	72.3	33.3
s	3.05	24.00	3.62	4.90	2.68	0.29	21.50	5.27	9.27	2.99
s_x	0.14	4.69	1.47	2.00	1.08	0.12	8.80	1.02	3.78	1.22

The mean of two consecutive determinations at the start of the experiment is compared with the mean of two consecutive determinations in the same animal six to eight days after the baseline was established. No injections of plasma were made.

s = Standard deviation

s_x = Standard error of the mean

weight was observed; this finding would be expected as the animals increased in size from normal growth. Figure 1 illustrates the serial values in a representative control rabbit.

The mean of two consecutive baseline determinations is recorded in Table I and compared with the mean for two consecutive determinations done between six and eight days after the second determination, the time interval at which a positive precipitin test would be expected were the animals injected with plasma. The statistical analysis is summarized in Table II and indicates that, in absolute values or in milliliters per kilogram of body weight on the day of the determination, no significant difference was found. This statement is true for plasma volume, thiocyanate space, total blood volume, hematocrit, and weight.

These data have subsequently been extended in a series of 45 animals in conjunction with other studies on normal values in rabbits (*7*).

General observations: Clinical serum sickness, manifested by erythema, edema, or both at the base of the ears, generalized subcutaneous edema and hyperirritability developed in 10 of the 11 animals in group 2, and in 11 of the 12 animals in group 3. Clinical signs usually began on the fifth or sixth day following the initial injection of plasma and usually lasted two or three days. The edema was most striking in the skin over the abdomen, but was not evident unless the fur had been clipped; the skin appeared pale and felt soggy, hot, and moist. Fever (rectal temperature over 40 C.) was not a constant feature of clinical serum sickness. The clinical severity of serum sickness bore no relation to the size of the sensitizing dose. Multiple injections produced no more severe reactions than did single injections. The subcutaneous route of sensitization was as effective as the intravenous route, but variations in absorption could not be detected or controlled.

TABLE II

Statistical significance of controls on methods (group 1)

	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit
	Kilo.	*ml.*	*ml.*	*ml.*	*Vol. %*
$\bar{d}$	+.11	+4.33	+10.00	+15.33	+.50
$S\bar{d}$	.05	13.60	5.30	9.35	1.81
t	2.20	.32	1.89	1.64	.28
		ml./Kilo.	*ml./Kilo.*	*ml./Kilo.*	
$\bar{d}$		−6.42	+1.67	+2.45	
$S\bar{d}$		3.67	1.90	3.58	
t		1.75	.88	.68	

No significant difference is noted between the baseline values and those obtained at the time of the expected response. The data are calculated in absolute values and in terms of weight on the day of the determination. The value of t was not considered significant if P was greater than .01.

Preliminary experiments: The alterations in plasma volume, thiocyanate space, total blood volume, and hematocrit which were observed in a representative rabbit sensitized with human plasma are illustrated in Figure 2. The changes in fluid distribution bore no direct relationship to the sensitizing dose of plasma, but paralleled the clinical severity of the disease.

In 10 of the 11 animals in group 2 increases in the *thiocyanate space* developed at some time during the period of observation. In eight animals two rises occurred, the first usually between the third and sixth days, and the second between the 11th and 20th days. The first rise usually coincided with the onset of the clinical manifestations; the second was usually marked by an increase in the subcutaneous edema. The changes in the thiocyanate space usually paralleled the weight changes, especially with the second reaction. Certain discrepancies were noted, however.

The most profound drop in *plasma volume* was noted in those animals which developed the severest clinical signs of serum sickness and the greatest and most prolonged increases in thiocyanate space. In seven animals there was no significant decrease in plasma volume, however.

In the four animals which showed a definite drop in plasma volume an appreciable decrease in the *total blood volume* also occurred. In four animals no changes in the total blood volume occurred during the experimental period.

In all the animals with severe early clinical manifestations of serum sickness a drop in *hematocrit* was associated with a rise in thiocyanate space and a decrease in plasma volume. In six animals no striking variations in the hematocrit occurred

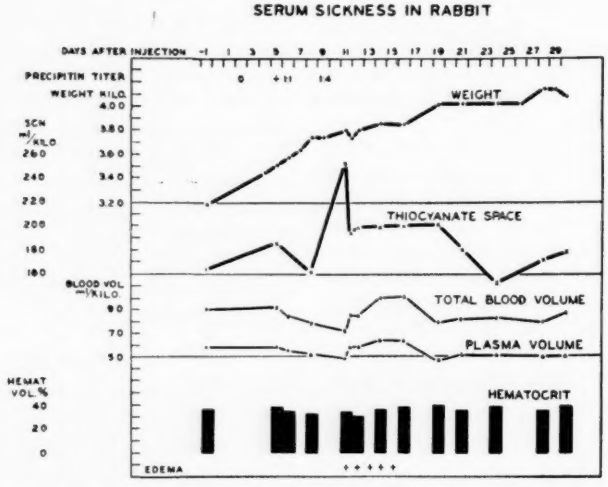

FIG. 2. FLUID SPACES IN A RABBIT (GROUP 2) GIVEN AN INTRAVENOUS INJECTION OF HUMAN PLASMA

Note the increase in thiocyanate space, which is out of proportion to the slight increase in weight, on the 11th day and the associated decrease in the plasma and total blood volume. On the following day, there was a marked decrease in the thiocyanate space, which was out of proportion to the decrease in weight. All the thiocyanate values from the 11th to the 21st day were elevated. After the 21st day, the variations in the thiocyanate space paralleled the weight changes and were not significant.

In the statistical calculations in the definitive experiment (group 3), Tables III and IV, two baseline values, two values obtained at the time precipitins appeared, and two values obtained at the termination of the experiment were used.

TABLE III

Alterations in fluid spaces in rabbits ill with serum sickness after an injection of human plasma (group 3)

Rabbit number	Baseline					Response*				
	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit
	Kilo.	*ml./Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*Vol. %*	*Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*ml./Kilo.*	*Vol. %*
18	2.10	228.5	46.0	75.5	39.0	2.10	322.5	48.0	74.0	35.0
19	2.14	235.5	48.5	80.0	40.0	2.14	242.5	47.0	75.5	37.5
20	1.99	246.5	48.5	79.5	39.0	2.14	282.5	39.5	61.5	35.7
21	1.87	245.5	45.0	75.5	40.5	1.86	296.0	39.0	62.5	37.5
22	1.74	289.5	48.5	78.5	37.5	1.96	273.0	44.0	68.5	35.2
23	1.87	266.0	45.5	74.0	38.5	2.04	313.5	40.0	62.0	36.0
24	1.76	229.5	50.0	80.0	37.0	2.05	281.5	35.0	54.5	36.0
25	2.10	212.5	51.0	84.0	39.5	2.30	265.0	39.0	62.0	36.7
26	2.22	183.0	54.0	88.5	39.0	2.24	225.5	40.5	62.5	35.7
27	1.76	215.0	63.0	108.0	42.0	1.93	280.0	47.0	78.0	40.0
28	1.96	202.5	52.0	85.5	39.5	2.07	307.5	40.5	63.5	36.0
29	1.85	220.5	54.0	92.0	41.0	1.90	311.0	48.5	78.0	38.5
Mean for group	1.95	231.2	50.5	83.4	39.4	2.06	283.4	42.3	66.9	36.7
s	0.17	28.60	4.95	9.47	1.38	0.13	28.00	4.40	8.44	1.48
s_x	0.05	8.21	1.45	2.73	0.40	0.04	8.42	1.27	2.54	0.42

* The mean of two consecutive determinations at the start of the experiment is compared with the mean of two consecutive determinations in the same rabbit done on the day precipitins to human plasma appeared and on the next day.
s = Standard deviation
s_x = Standard error of the mean

with the initial increase in thiocyanate space. None of the animals revealed a decrease in hematocrit in association with the second rise in thiocyanate space.

TABLE IV

Statistical significance of alterations in fluid spaces in rabbits ill with serum sickness (group 3)

	Weight	Thiocyanate space	Plasma volume	Total blood volume	Hematocrit
	Kilo.	*ml.*	*ml.*	*ml.*	*Vol. %*
$\bar{d}$	+.11	+126.00	−11.08	−24.67	−2.73
$S\bar{d}$	.03	21.40	2.81	4.57	.23
t	3.70	5.89	3.94	5.40	11.87
		*ml./Kilo.**	*ml./Kilo.*	*ml./Kilo.*	
$\bar{d}$		+51.30	−8.50	−16.50	
$S\bar{d}$		9.85	1.49	2.60	
t		5.21	5.70	6.35	
		ml./Kilo.†	*ml./Kilo.*	*ml./Kilo.*	
$\bar{d}$		+74.16	−6.25	−12.25	
$S\bar{d}$		9.98	1.23	2.35	
t		7.43	5.08	5.21	

* Calculated in terms of weight on the day of determination.
† Calculated in terms of weight on the day of the first baseline.
The data all show statistically significant differences between the baseline values and those obtained at the time of the humoral antibody response. The value of t was considered significant if P was <.01.

In all 11 animals positive *precipitin tests* to human plasma developed between the third and the 12th days following the initial injection of plasma. In eight animals the thiocyanate space decreased and the clinical symptoms and signs subsided as the precipitin tests became positive. In all animals except one the precipitin tests remained positive throughout the period of observation. In this animal, which was followed longer than the others, the precipitin test was negative after the 28th day.

Definitive experiment: The alterations in plasma volume, thiocyanate space, total blood volume, hematocrit and body weight are summarized in Table III.

The statistical analyses are summarized in Table IV.

A statistically significant increase in *thiocyanate space* was noted in each animal at the time the humoral antibody was first detected. In all instances a return to baseline values had occurred by the 20th day after injection of the sensitizing dose. A significant increase in body *weight* oc-

curred during the period of alteration in thiocyanate space. The body weight continued to increase from normal growth after the animal recovered from serum disease.

In contrast to the increase in thiocyanate space and weight, a statistically significant decrease in *plasma volume, total blood volume* and *hematocrit* occurred in all animals.

All of the data are statistically significant, whether the calculations are based on the comparison of *1*) absolute values; *2*) values based on milliliters per kilogram of weight on the day of the determination; or *3*) values based on milliliters per kilogram of weight on the day of the first baseline measurement.

In order to determine whether these changes were reversible, the mean of two consecutive determinations at the termination of the experiment was compared with the mean baseline values in each animal. The mean differences for the body weight, plasma volume, total blood volume, and the thiocyanate space were not significant. The decrease in the hematocrit at this time was significant. These data are not included in the tables.

In all 12 animals positive *precipitin tests* to human plasma developed between the fourth to the eighth day after injection. The distribution was as follows:

	Onset of positive precipitin tests				
Days after injection	4	5	6	7	8
Number of rabbits	2	1	2	4	3

DISCUSSION

The experiments indicate that immune reactions may be associated with alterations in the blood volume and the fluid space which is available for the diffusion of thiocyanate ion. The maximum alterations in rabbits sensitized with pooled human plasma occurred at the time circulating humoral antibodies (precipitins) appeared in the serum. This finding correlates well with the previous clinical observation that the symptoms of serum sickness begin to disappear with the appearance of precipitins (3).

The greatest change was observed in the thiocyanate space. Alterations in the plasma volume and the hematocrit were less pronounced in magnitude and were more transient, lasting only one to three days as compared with the changes in thiocyanate space which occasionally persisted into the second week. The weight of the animals tended to increase with the increase in thiocyanate space, though the changes could not always be correlated closely. Apparently the normal distribution of fluid in the body was altered as the result of an increase in permeability of membranes which was completely reversible.

Methods: The chemical methods were reliable since the results in the control animals (group 1) were consistently reproducible. The possibility that the dye might act as an antigen, producing serum sickness, or as a haptene, forming a new compound which might induce an auto-antibody response, was ruled out by the preliminary experiments.

The variability between individual animals in the experimental group (group 3) was controlled by determining baseline values on each rabbit and comparing these values with the response in the same animal for the statistical analysis. The effect of growth and of variations in food and water intake was apparently of little significance in this experiment, since the data were found to be statistically significant whether calculated in terms of absolute values or in relation to weight at the time of the first baseline determination or on the day of the immunologic and clinical response.

Though whole plasma is a complex of many protein fractions, each of which might induce its own slightly different immunologic response, it was felt that a mixed antigen might more closely simulate a natural infection in which multiple antigens are liberated by the infecting organism. Anatomic studies have indicated that the body responds at different times after the injection of albumin or globulin fractions derived from the same plasma (5). The two peaks in the thiocyanate space observed in the preliminary experiments may be due to this difference.

Permeability of membranes: The present experiment offers a logical explanation for the phenomenon of retention of salt and water; if the permeability of membranes to ions is increased, these ions could diffuse from blood into interstitial spaces and from interstitial fluid spaces into cells. The

resultant hypochloremia would decrease the renal excretion of salt and water. The administration of saline would increase the edema, until restoration of the integrity of membranes corrected the alterations in the internal environment. It is not clear whether the physiologic alteration is confined solely to vascular walls, as pathologic experiments might suggest, or whether cellular membranes are also affected. Von Pirquet suggested that the edema was interstitial, but the data in the present experiments may indicate that some of the change occurs within cells (8).

Capillary permeability: The alterations in the vascular tree can be visualized as a series of pores in the capillary wall whose size is related to the clinical severity of the disease. In mild instances of the disease water and ions which normally traverse the membrane would leave the blood more readily. Larger molecules such as protein would be lost in more severe instances and red blood cells would pass out of the vascular tree only in the most severe reactions. Petechial hemorrhages in organs have been recognized for years as a manifestation of anaphylactic reactions.

The loss of water and ions from the vascular tree into tissues is compatible with the observations on retention of water, sodium and chloride during pneumonia and serum disease (1, 2). The permeability of the vascular tree to some colloids must have been increased because the decrease in plasma volume was concomitant with the increase in thiocyanate space. The colloids lost from the circulation to the interstitial spaces would tend to retain water extravascularly until they had been reabsorbed, presumably through the lymphatics, broken down in the tissues, or until other homeostatic mechanisms had been operative.

It is probable that the amount of protein lost from the vascular tree during serum disease is not great. The dye is known to be bound to albumin, one of the smaller protein molecules of the blood. Any great loss of albumin into interstitial spaces would reduce the amount of circulating dye and hence lead to a larger value for plasma volume by the chemical method; a loss in blood volume of any magnitude would thus be obscured. If protein was lost from the vascular tree, the defect was detected only with the first rise in thiocyanate space and was transient.

The drop in hematocrit though slight may indicate a loss of red blood cells from the vascular compartment. The drop was apparently not due to repeated bleeding since the hematocrit remained constant in group 1 in which more determinations were done in the same interval of time. No jaundice was noted clinically and the serum was never icteric, hence it seems unlikely that the red cells were hemolyzed. It is possible that the red cells were reduced in size or that they were pooled in some portion of the vascular tree which was not measured by the techniques used.

Cellular permeability: It is not known whether the thiocyanate ion normally diffuses into cells or not. The increase in thiocyanate space therefore could be due to *1*) interstitial edema, *2*) increase in permeability of cells to the thiocyanate ion, *3*) destruction of tissue cells with liberation of cellular water, or *4*) dehydration of cells with a shift of intracellular fluid into the interstitial spaces. The fact that some animals showed a sudden increase in thiocyanate space without an increase in body weight would favor the thesis that some redistribution of fluid had occurred whether or not additional fluid ingested in food or water was retained. The hypothesis that tissue cells were destroyed, liberating additional water and making it available for dilution of thiocyanate ion, does not seem likely. The rapidity of the increase in the thiocyanate space and its reversibility makes this explanation appear highly improbable. Such a sudden increase in thiocyanate space without an increase in body weight would have to mean that the associated tissue destruction exceeded the amount of interstitial edema. Furthermore, preliminary nitrogen balance studies in other rabbits have disclosed no marked increase in the excretion of urinary nitrogen, and presumably in cell destruction, during clinical serum sickness.

Mechanisms: It is not known whether the antigen in serum disease attaches solely to the vascular wall or to tissue cells outside the vascular system as well. As far as the effect on the circulation is concerned, it would be immaterial whether the loss of substances from the vascular tree is attributable to an abnormal passage of fluid between the endothelial cells, as would occur if the permeability of the intercellular cement substance were decreased, or whether the permeability of the

cells themselves is abnormally increased, so that the fluids pass through the cells into the interstitial spaces.

The participation of cells in immune reactions is recognized clinically in allergic states; the deleterious antigen-antibody reaction is assumed to occur on the cell surface or perhaps within the cell. It has been postulated that histamine or some similar substance is released during the interaction of antigens with antibodies. Histamine will alter the local permeability of capillary walls. The liberation at the cell wall of some substance which would increase the permeability of the cell membrane would enhance the transmission of molecules, including the thiocyanate ion, across the surface. Such a hypothesis is of more than academic interest, since it is known that intracellular enzyme systems are affected by the extent of hydration of cells and by the concentration of ions. An increased cellular content of ions could be proved by isotopic techniques which could measure not only the diffusion of labelled thiocyanate but the relative movement of radioactive sodium, potassium, and chloride ions across cell membranes.

Relation to infections: The present findings indicate that the physiologic alterations which may result from antigen-antibody reactions are more profound than has been recognized in the past. The physiologic changes deserve more attention in the therapy of infectious diseases. In infections, the invading micro-organisms multiply within the host and release or secrete multiple antigens. With organisms known to locate intracellularly during some phase of the disease, as is true with the viruses and rickettsias, antigens are produced within the cell. Those antigens which could readily diffuse out would produce predominantly humoral antigen-antibody reactions; those antigens which would be retained within cells would be more likely to induce intracellular antigen-antibody reactions and hence increase the extent of the permeability of cellular walls. A most profound alteration in functions of the body could thus be produced. The possible application of antihistaminic drugs or adrenal cortical hormones to therapy of infections with an allergic element in the host response is based on such alterations in function.

SUMMARY

1. Rabbits sensitized by the intravenous injection of pooled human plasma developed clinical edema with significant increases in weight and in the fluid space available for dilution of thiocyanate ions.

2. Significant decreases in the plasma volume, total blood volume and hematocrit were observed transiently during the most marked alteration of thiocyanate space.

3. The changes were interpreted as indicating an increase in permeability of the vascular tree, probably in the capillary wall, so that crystalloids, colloids, and red blood cells are progressively lost from the circulation as the magnitude of the defect increases with increase in the clinical severity of the disease.

4. Occasional discrepancies observed between the changes in thiocyanate space and in weight suggest that cellular permeability is probably also increased to water and ions.

5. The physiologic changes occurred at the time humoral antibodies (precipitins) appeared, suggesting that the alterations may have been due directly or indirectly to an antigen-antibody reaction.

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INTRACELLULAR CATION EXCHANGES IN METABOLIC ALKALOSIS [1]

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It is now well known that disturbances in the electrolyte patterns of the intracellular fluid, as well as in those of the extracellular fluid, may play a significant role in many disease states. Darrow and associates (1) have shown in experimental animals that one pattern of associated abnormalities in the two fluid phases involves intracellular potassium deficit plus sodium excess and extracellular bicarbonate excess plus deficit of chloride. Since then evidence of this relationship has been found in a variety of clinical conditions including congenital alkalosis, vomiting, the postoperative state, and syndromes of hyperadrenocortical function such as Cushing's disease and ACTH administration. This evidence has been obtained by utilization of the balance technic during replacement therapy (2–8), by study of the distribution of radioactive sodium (9), and by tissue analyses of skeletal muscle (10) and of erythrocytes (9, 11). These observations have suggested that both low potassium intake and hyperadrenocortical function are implicated in the etiology of this disturbance. In addition to these two etiological factors a third has been postulated, namely, impaired renal conservation of the potassium ion (12).

The purpose of the present study is to examine, in a group of patients with metabolic alkalosis and hypokaliemia, *1*) the direction and magnitude of intracellular electrolyte abnormalities as revealed by the balance technic, *2*) the relation of these abnormalities to the extracellular hypochloremia and elevation of bicarbonate, and *3*) the relative etiological roles of gastrointestinal fluid loss, low potassium intake, intrinsic renal dysfunction, and abnormal adrenocortical activity.

EXPERIMENTAL MATERIAL AND METHODS [5]

Ten patients were studied by the balance technic, eight of whom were followed during fluid replacement therapy and correction of the alkalosis. The diagnoses of these ten patients and their pertinent clinical conditions are listed in Table I.

Fluids and food of known composition were administered intravenously or orally. All excreta were collected and analysed with the exception of formed stools which usually contain insignificant amounts of electrolytes when no salts have been administered by mouth. Sodium and potassium were determined in serum, urine, and nitric acid digests of stools and foods by means of a Barclay internal standard flame photometer according to the methods of Wallace, Holliday, Cushman and Elkinton (13). Chloride was determined in serum by the method of Eisenman (14), in urine by the Volhard-Harvey method (15); nitrogen was determined by micro- or macro-Kjeldahl. Total CO_2 content of serum and blood was measured by the method of Van Slyke and Stadie (16), and pH determinations were made with the glass electrode or colorimetrically. Creatinine in urine was determined according to the method of Brod and Sirota (17) and in serum according to the method of Bonsnes and Taussky (18). Inulin determinations in serum and urine were made according to the method of Harrison (19); spaces and clearances being determined according to the formulae, $\frac{In-Out}{P}$ and $\frac{UV}{P}$, respectively. The method for the determination of urinary 17-ketosteroids was modified from that of Holtorff and Koch (20).[6]

The derived data were calculated according to previous methods (21). The fundamental assumption involved is the equating of the extracellular fluid with the chloride space; this assumption is discussed below. The change in chloride space is calculated from the change in concentration of chloride in serum (corrected for serum water and the Donnan equilibrium), the balance of chloride, and an assumed initial or final volume for the chloride

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[6] The determinations of urinary 17-ketosteroids were done in the Endocrine Laboratory under the direction of Dr. F. Curtis Dohan.

TABLE I

Diagnosis, pertinent history, and status of renal and adrenocortical function

Patient	Age	Sex	Color	Diagnosis	Pertinent history prior to study	Renal factors				Adrenocortical function		
						BUN	Clearance			17-keto-steroids	Total N balance	Remarks
							Creatinine	Inulin	PAH			
	years					*mg. per cent*	*ml. per min.*	*ml. per min.*	*ml. per min.*	*mg. per day*		
Group I: Low-K intake												
W. L.	28	M	N	Psychoneurosis—starvation	Very little K-containing food or fluids for 4 weeks. No vomiting or diarrhea	16–12	—	65	270	4.9	Pos.	Clinical diagnosis of Addison's disease ruled out by Na conservation on 6 days low Na intake
F. G.	33	F	W	Anorexia nervosa	Semi-starvation for 5 weeks	8–7	22–46*	—	—	1.6	Pos.	
Group II: Low-K intake plus gastrointestinal fluid loss												
A. K.	57	F	W	Chronic ulcerative colitis	Maintained on low K, Amigen solution i.v., for 4 weeks; extensive diarrhea	14	86†	106	630	7.3	Neg.	
N. D.	53	F	W	Hyperthyroidism	Vomiting, 8 weeks	26–28	27–36*	—	—	8.5	Neg.	
W. H.	6 wks.	M	W	Congenital pyloric stenosis	Vomiting, 11 days	—	—	—	—	—	Pos.	
H. H.	67	M	W	Cholecystitis, cholecystectomy	Postop. pyloric ileus, 12 days	76–33	—	—	—	14.0	Neg.	
T. W.	59	M	W	Chronic hepatitis with massive liver necrosis	Gastric ileus, 10 days	60–92	—	—	—	—	—	
A. J.	71	F	N	Carcinoma of colon with colostomy	Vomiting and colostomy drainage, 8 weeks	8	—	—	—	—	—	
Group III: Adrenocortical hyperfunction												
M. F.	5	M	W	Dermatomyositis	Large dosage of ACTH while on K-free fluids, 4 days	—	—	—	—	—	Neg.	17-Ketosteroids elevated during previous course of ACTH
J. C.	46	F	W	Carcinoma of adrenal cortex with adrenogenital syndrome	Hirsutism and male habitus, 2 years; amenorrhea, 1 year; tumor in left flank	8–11	55–113*	—	—	80–85	Pos.	No evidence of increased 11-oxysteroid activity

* 24-hour clearance. † Acute clearance measured simultaneously with that of inulin.

TABLE II

External exchanges of water, electrolytes and nitrogen

Patient	Period		Main fluid therapy	Average daily intake					Average daily output										Total period balance*			
									Urine					G.I. fluid†								
	Dates	Duration		H_2O	Cl	Na	K	N	Vol.	Cl	Na	K	N	Vol.	Cl	Na	K	N	Cl	Na	K	N
		days		*ml.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*ml.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*ml.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*
W. L.	3/15–17	2	NaCl+milk	4,050	124	110	70	8.5	2,945	23	41	2	4.3						+200	+136	+135	+ 8.4
	3/17–21	4	$KHPO_4$−KCl−NaCl‡	4,739	168	113	159	9.5	3,875	46	90	8	5.5						+482	+ 80	+604	+16.0
	3/21–25	4	Milk	3,713	62	46	85	10.4	3,455	61	107	44	8.8	75	0	2	5	0.4	− 2	−262	+145	+ 4.7
F. G.	3/8 –11	3	NaCl+Amigen	1,938	163	200	1	7.9	676	7	6	5	1.7						+467	+576	− 10	+18.4
	3/11–21	6	Soft food	2,900	13	19	72	4.5	887	12	39	4	1.5						+ 5	−128	+407	+17.9
A. K.	9/16–20	4	NH_4Cl§+Amigen	2,913	174	84	3	8.4	1,100	34	35	19	11.2	131	5	2	2	0.2	+537	+181	− 73	−12.3
	9/20–24	4	KCl‖+Amigen	3,063	148	36	135	4.5	575	46	25	41	2.6	941¶	42	36	32	6.9	+234	−108	+330	−20.1
	9/24–28	4	Protenum	1,956	54	53	65	11.5	554	28	20	23	3.9	455	30	29	25	4.1	− 20	+ 5	+ 70	+14.0
N. D.	7/8 –11	3	$KHPO_4$−KCl−NaCl‡+milk	4,428	201	193	139	9.3	2,618	125	159	65	11.9						+225	+ 96	+224	− 7.9
	7/11–13	2	KCl+milk	3,970	180	97	174	10.4	2,340	69	92	72	11.8	265	17	11	7	1.9	+187	− 16	+191	− 6.5
W. H.	8/24–27	3	KCl‖+milk	1,190	55	50	18	2.3	502	2	17	2	0.3	272	22	8	7	—	+ 94	+ 75	+ 25	+ 2.3
	8/27–31	4	Milk−postop.	672	22	18	17	2.4	145	1	7	2	0.3	48	4	2	1	—	+ 68	+ 36	+ 53	+ 7.6
H. H.	1/19–21	2	NH_4Cl§+KCl	4,163	474	307	40	0	1,150	7	77	55	13.9	1,305	193	194	12	3.3	+547	+ 68	− 54	−34.3
	1/21–24	3	NaCl+food	2,667	119	118	14	3.2	1,108	9	49	34	15.5	383	19	20	5	0.9	+269	+150	− 73	−39.7
	1/24–28	4	KCl‖+food	3,253	164	138	51	3.0	753	11	56	29	9.5	756	87	18	10	2.1	+261	+247	+ 49	−34.1
T. W.	2/7 –10	3	NaCl	3,337	323	336	0	—	404	1	5	11	—	2,500	186	70	51	—	+406	+783	−187	—
	2/10–12	2	NaCl+KCl‖	4,585	307	235	92	—	150	4	7	2	—	2,250	226	67	16	—	+156	+323	+141	—
A. J.	2/21–22	1	NaCl+KCl‖+milk	3,975	348	—	128	—	800	142	—	23	—	200	25	—	2	—	+179	—	+103	—
M. F.	6/7 –9	2	$KHPO_4$−KCl−NaCl‡	735	41	20	46	1.9	180	3	1	4	3.7	350††	3	1	4	3.7	+ 16	− 60	+ 85	− 2.8
	6/9 –13	4	$KHPO_4$−KCl−NaCl‡	2,394	62	36	64	6.9	1,386	43	95	25	7.3						+ 71	−240	+156	− 1.9
J. C.	1/30–2/2	3	Milk, Low K milk	3,833	57	52	37	7.6	2,142	102	120	34	8.2						−146	−211	+ 9	− 1.8
	2/2 –4	2	NH_4Cl**+Low K milk	3,500	118	12	13	9.9	925	21	16	13	7.0						+192	− 11	+ 1	+ 5.9
	2/4 –7‡‡	3	KCl**+milk	4,117	107	45	130	10.3	1,138±	36±	11±	17±	8.0±						+213±	+102±	+199±	+ 4.1±

* Balances of Cl and Na corrected for small quantities lost in serum drawn for analysis.
† Includes vomitus, gastric drainage, and unformed stools.
‡ Given as an intravenous solution containing Cl^- 112, HPO_3^{--} 42, Na^+ 74, and K^+ 80 m.eq. per l., respectively; "$KHPO_4$" indicates a buffered solution of K_2HPO_4 plus KH_2PO_4.
§ Given intravenously as a 2 per cent solution.
‖ Given intravenously as 0.6 per cent solution.
¶ Includes urine mixed with feces.
** Given by mouth.
†† Pleural fluid.
‡‡ Urine lost on first day of this period.
Balance data are expressed per individual period rather than cumulatively.

TABLE III

Analyses of blood and serum, and changes in the distribution of sodium and potassium between the phases of body fluid

Patient	Time		Main fluid therapy	Weight	Serum					Blood pH	Extracellular volume†	Change in						
												Extracellular		Intracellular				
	Date	Period duration			CO_2	Cl	Na	K	H_2O			Na	K	Na	K*	Na	K*	K*
		days		*kg.*	*m.eq. per l.*	*m.eq. per l.*	*m.eq. per l.*	*m.eq. per l.*	*gm. per l.*		*l.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq. per kg.*
W. L.	3/15			48.0	45.0	79	137	2.4	939		5.6							
	3/17	2	NaCl+milk	—	40.1	88	142	2.5	943	7.49	7.2	+250	+ 5	−114	+120			
	3/21	4	$KHPO_4$−KCl−NaCl	50.9	30.6	101	140	4.0	929		10.3‖	+440	+23	−360	+543	↓	↓	↓
	3/25	4	Milk	49.9	27.9	104	141	5.7	928		(10.0)	− 30	+18	−232	+115	−706	+778	+15.6
F. G.	3/8			—	45.5	74	127	2.8	930‡		15.9							
	3/11	3	NaCl+Amigen	—	33.8	98	137	2.4			16.3	+210	− 6	+366	− 48			
	3/21	6	Soft food	39.6	31.5	100	132	4.2			(16.0)	−120	+28	− 8	+336	− 8	+336	+ 8.5
A. K.	9/16			—	45.9	80	136	2.3	943		10.0							
	9/20	4	NH_4Cl+Amigen	—	27.9	93	136	2.3	945		13.7	+505	+ 9	−324	− 52			
	9/24	4	KCl+Amigen	—	23.8	97	131	5.0	938		15.2	+145	+44	−253	+335	↓	↓	↓
	9/28	4	Protenum	—	28.4	94	133	4.2	947		(15.6)¶	+ 55	+43	− 50	+ 46	−627	+329	+ 5.5**
N. D.	7/8			—	38.6	93	147	2.7	918	7.43	6.9							
	7/11	3	$KHPO_4$−KCl−NaCl+milk	—	38.7	98	149	4.0	923		8.6	+271	+15	−175	+228	↓	↓	↓
	7/13	2	KCl+milk	42.8	30.6	106	148	3.7	917		(9.4)	+122	+ 1	−138	+205	−313	+433	+10.1
W. H.	8/24			3.5	52.8	62	131	3.2	930‡		0.6							
	8/27	3	KCl+milk	—	40.1	95	138	3.3	930‡		1.3	+ 98	+ 2	− 22	+ 17	↓	↓	↓
	8/31	4	Milk	3.6	23.0	102	145	5.9	930‡		(1.8)	+ 83	+ 6	− 47	+ 29	− 69	+ 46	+12.7
H. H.	1/19			—	44.6	78	140	3.9			8.5							
	1/21	2	NH_4Cl+KCl	—	36.9	89	139	4.0			13.0	+630	+19	−562	+ 4	↓	↓	↓
	1/24	3	NaCl+food	68.6	31.0	93	132	3.6	934§		14.9	+165	+ 2	− 5	+ 21			
	1/28	4	KCl+food	—	30.2	100	142	4.8			(16.2)	+320	+14	−173	+117	−740	+142	+ 2.1
T. W.	2/7			—	25.7	100	140	3.9	935§		(10.0)							
	2/10	3	NaCl	—	40.0	88	152	2.8		7.57	15.5	+980	+ 3	−197	−190††			
	2/12	2	KCl+NaCl	—	38.7	77	151	3.4			19.4	+570	+23	−247	+118††	−247	+118††	—
A. J.	2/21			—	38.3	89	146	1.8	949									
	2/22	1	KCl+NaCl	—	41.4	88	143	2.2	946			—	—	—	—	—	—	—
M. F.	6/7			—	40.0	88	137	1.6	934	7.48	6.2							
	6/9	2	$KHPO_4$−KCl−NaCl	63.5	45.0	82	140	3.4			6.9	+119	+14	−179	+ 78	↓	↓	↓
	6/13	4	$KHPO_4$−KCl−NaCl	56.0	26.0	95	134	4.2	928		(6.5)	− 97	+ 3	−143	+157	−322	+235	+ 9.1
J. C.	1/30			61.3	31.0	105	142	2.9	937§		(15.0)							
	2/2	3	Milk, low-K milk	58.7	32.9	102	144	2.7			14.1	−100	− 6	−111	+ 20	↓	↓	
	2/4	2	NH_4Cl+low-K milk	58.7	29.3	104	141	2.7			15.5	+160	+ 4	−171	− 17	−282	+ 3	—
	2/7	3	KCl+milk	59.3	27.0	106	143	4.3	937§		—	—	—	—	—	—	—	—

* In excess of nitrogen.
† Calculated by changes in chloride balance forward or backward from an assumed extracellular volume (designated by parentheses).
‡ Assumed.
§ Calculated from the concentration of serum protein.
‖ Inulin space measured at this time equalled 6.4 l.
¶ Inulin space measured at this time equalled 8.4 l.
** Approximated.
†† Total intracellular K, not K in excess of N.
Except where indicated by arrows, derived data are expressed per individual period rather than cumulatively.
Time as designated by date indicates end of period at which time serum analyses were made, balances for the period were determined, and exchanges calculated.

space. The inulin space was determined in two patients at the end of the period of study, but for reasons outlined in the *Discussion* were not used in the calculation of change in chloride space. From the values obtained for the volume of extracellular fluid and from the external balances of sodium and potassium, the exchanges of these two ions between the extra- and intracellular phases of the total body fluids are calculated. The results of this latter calculation are little affected by considerable variation in the volume of extracellular fluid as originally assumed or measured.

RESULTS

The results are presented in Tables I to IV, and in Figure 1. The clinical diagnoses of the patients studied are shown in Table I.

Intracellular cation exchanges. In all but one (H. H.) of the ten patients potassium was retained when administered (Table II). In those patients in whom it was possible to make the calculation, the retained potassium entered the intracellular phase in excess of nitrogen, in amounts up to 15.6 m.eq. per kilogram of body weight (Table III). During treatment, in seven out of ten patients, the balance of chloride was markedly positive in relation to that of sodium (Table II). On the assumption that chloride exchanges were extracellular, it is calculated that large amounts of sodium left the intracellular phase in these patients (Table III). In some of the patients (W. L., N. D., and M. F.) the increment of potassium and the decrement of sodium were of similar magnitude; in others, more sodium was lost from the cells than potassium was gained (Table III). Only two patients were not shown to lose intracellular sodium during treatment, and in one of these, A. J., the sodium exchanges were not measured.

Relation of intracellular cation exchanges to extracellular hypochloremia and bicarbonate elevation. In all the cases except J. C. the elevated bicarbonate concentration in serum was associated with a lowered chloride concentration; and in all the cases during treatment the latter rose as the former fell (Table III). The high levels of bicarbonate were not associated with any marked degree of hypernatremia; during treatment in all cases except M. F. the extracellular sodium concentration remained unchanged or rose. During correction of this hypochloremic alkalosis in eight of ten cases reported the associated intracellular cation exchanges were as follows. In four cases

TABLE IV

Relation of observed chloride exchanges to increments of chloride calculated as necessary to reduce serum CO_2 concentration to 31 m.eq. per l. (During early periods before response of alkalosis to therapy)

Patient	Period	Serum		Chloride			
		CO_2	Cl	Given	Retained	Excreted in urine	Increment to lower CO_2*
	days	*m.eq. per l.*	*m.eq. per l.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*
W. L.	0	45.0	79				
	2	40.1	88	247	200	45	66
H. H.	0	44.6	78				
	2	36.9	89	948	547	13	77
M. F.	0	40.0	88				
	2	45.0	82	82	16	5	97
N. D.	0	38.6	93				
	3	38.7	98	603	225	374	67
W. H.	0	52.8	62				
	3	40.1	95	165	96	72	12
A. J.	0	38.3	89				
	1	41.4	88	348	179	142	124
J. C.	0	31.0	105				
	3	32.9	102	170	−146	307	27

* Calculated from the formula, $E_2 \times ([CO_2]_{s2} - 31)$, where E_2 = extracellular volume at end of the period as derived from an assumed value and the balance of chloride, in liters; $[CO_2]_{s2}$ = concentration of bicarbonate in serum at end of period in m.eq. per l.

(W. L., N. D., W. H., and M. F.) to whom potassium was administered from the start of therapy, intracellular potassium increased and intracellular sodium decreased as the bicarbonate concentration fell. In three others (A. K., H. H., J. C.) to whom NH_4Cl was administered first, the bicarbonate concentration fell to normal levels and sodium was extruded from the intracellular phase in the absence of any significant uptake of potassium. In two of these three cases (A. K. and J. C.) "excess" cellular potassium was subsequently taken up when the ion was administered; in the third, H. H., potassium was not retained when given. In the eighth case, F. G., the fall in bicarbonate concentration occurred in conjunction with a rise in intracellular sodium followed by a rise in intracellular potassium. The dissociation in some of the cases of the uptake of intracellular potassium from the correction of the elevated bicarbonate concentration and its frequent association with the extrusion of intracellular sodium, as shown in three different cases, is illustrated in Figure 1.

Etiology. The etiological factors responsible for these electrolyte abnormalities are less readily analysed since in nine of the ten cases no observations were made during the development of the alkalosis. A rough etiological classification of the cases has been attempted in Table I. (*a*) *Low-K intake:* Two cases (Group I) gave a history of a low-K intake without any concomitant gastrointestinal fluid loss. Both of these cases showed a maximal renal response for conserving potassium at the time the studies were initiated (see below), and neither case showed any evidence of in-

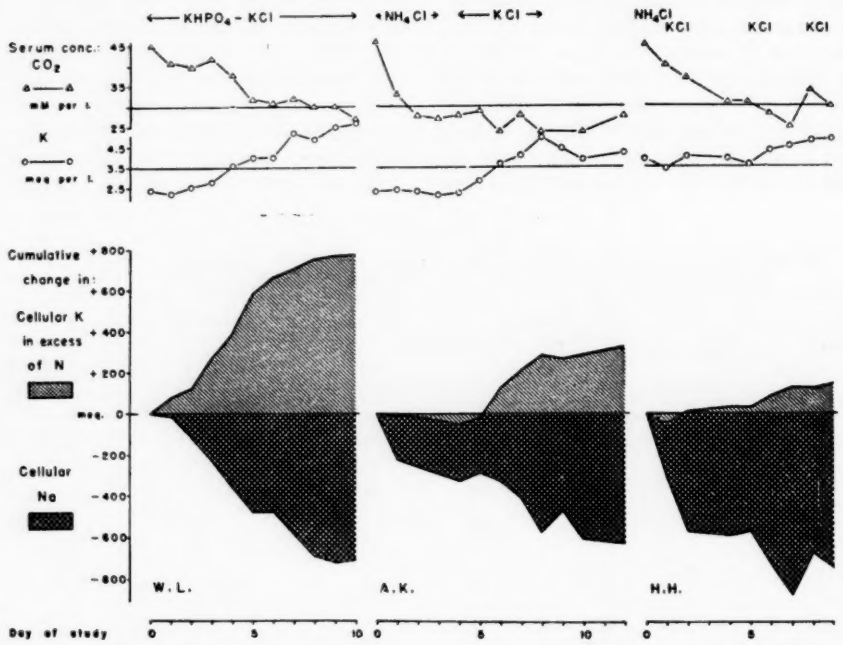

FIG. 1. RELATION OF CHANGES IN SERUM LEVELS OF CARBON DIOXIDE AND POTASSIUM TO CUMULATIVE CHANGES IN INTRACELLULAR CATIONS, DURING TREATMENT OF METABOLIC ALKALOSIS; THREE TYPES OF RESPONSE

In Patient W. L. intracellular potassium increased and sodium decreased simultaneously with the return of serum CO_2 and K to normal levels. In Patient A. K. during administration of NH_4Cl the serum CO_2 level returned to normal coincidently with a loss of cellular sodium and prior to an increase in intracellular potassium. In Patient H. H. the serum CO_2 level returned to normal in association with a loss of intracellular sodium and without any significant subsequent uptake of cellular potassium.

creased adrenocortical activity. (*b*) *Gastrointestinal fluid loss:* Six cases (Group II) were known to have extensive losses of gastrointestinal fluid containing potassium in addition to giving a history suggestive of a low-K intake. Since, with the exception of A. K., the gastrointestinal fluid lost included gastric fluid, probably greater amounts of chloride were removed than of sodium. (*c*) *Adrenocortical hyperfunction:* In Group III are listed two cases in which this factor apparently played a major etiological role. In M. F. the administration of large doses of ACTH, in conjunction with a low intake of potassium, immediately preceded the onset of prostration associated with a severe alkalosis and hypokaliemia. This patient had been shown during a previous course of ACTH to have an adrenocortical response as evidenced by a fall in circulating eosinophils, a rise in 17-ketosteroid excretion, and retention of sodium (22). In the other case, J. C., who had a typical adrenogenital syndrome due to carcinoma of the adrenal cortex, hyperfunction of the gland was the only factor that could be adduced to account for the mild alkalosis and hypokaliemia found. In none of the other cases was there unequivocal evidence of adrenocortical hyperfunction occurring as an etiological factor although thorough and systematic studies of this aspect of the problem were not undertaken. In five of the other eight cases the excretion rate of 17-ketosteroids was determined and was found to be normal or low (Table I). A negative nitrogen balance which might indicate adrenogenital hyperfunction was found in three of these cases during treatment. Of these A. K. had an extensive loss of nitrogen from the bowel, N. D. was thyrotoxic, and H. H. was in the immediate postoperative period. It is therefore difficult to use this observation as evidence of primary adrenocortical hyperfunction. (*d*) *Renal dysfunction:* kidney function was not systematically studied in this series of cases. Certain observations in some of the cases, however, are worthy of note (Table I). Five cases did not have azotemia; two of these, W. L. and F. G., were patients who apparently developed the alkalosis and potassium deficiency as the result of a low-K intake alone. During the first few days of study at the height of the alkalosis both of these patients excreted such extremely small amounts of potassium that the U/P ratio was less than 1.0. Conservation of potassium, therefore, was maximal at that time, despite the evidence from clearances of creatinine and inulin that glomerular filtration was impaired.

DISCUSSION

The observations made in this study confirm the hypothesis that extensive disturbances of intracellular electrolyte exchanges usually occur in states of metabolic alkalosis. Intracellular deficit of potassium, as measured by uptake of the ion during treatment, was certainly present in most of the cases. In Patient W. L. the deficit measured in this way reached a maximum of 15.6 m.eq. per kilogram of body weight, and is of the same magnitude as the largest deficits reported in infant diarrhea (23) or diabetic acidosis (24). If it is assumed that all of this potassium came from skeletal muscle and that this tissue constitutes 40 per cent of the body weight, the deficiency would be equivalent to 39 m. eq. per kilogram of wet muscle and would approximate 40 to 50 per cent of the potassium in the intracellular phase. Such a deficit is quite comparable to those produced experimentally in rats by Darrow (1) and exceeds those found in muscle biopsies taken from clinical cases by Mudge and Vislocky (10).

The demonstration of an excess of sodium in the intracellular phase is less certain. As calculated on the basis of the assumption that the chloride exchanges were entirely extracellular, the intracellular sodium exchange during treatment was negative in all but one of the nine cases in which it was measured. In four of the cases the size of the decrement of cellular sodium was sufficiently close to that of the increment of potassium to suggest that the former cation was being substituted for by the latter. In the other four cases the loss of cellular sodium so far exceeded the uptake of potassium as to obviate any such relationship. The validity of the assumption, however, on which these exchanges of intracellular sodium are calculated, must be questioned. Certainly in most of the cases during treatment the external balance of chloride was much greater in the positive direction than that of sodium. These balances are observed, not derived, data, and would not be invalidated by any unmeasured loss of salt in sweat in which chloride and sodium are present in approximately equal concentrations. The differential secretion of Cl into gastric fluid which was not lost from the body

and therefore not included in the measured balance, conceivably might be another source of error. The magnitude of the discrepancies between Cl and Na renders this explanation improbable; it is unlikely that, during the course of study in some of the patients, 5 or 6 l. of unreabsorbed gastric juice accumulated undetected. The difference between the external balances of these two ions would appear to require some other explanation. An alternative to the interpretation that these disparate balances represented loss of sodium from the cells is that chloride entered the intracellular phase. Physiological evidence for such a phenomenon is not lacking. Conway's (25) theory calls for a small amount of intracellular chloride to balance the extracellular potassium, and Dean (26), Wilde (27), and Burch and co-workers (28) have presented evidence that under certain circumstances chloride may enter the cells of tissues other than gastric mucosa, testes, and ovaries where chloride is a normal intracellular constituent. Evidence in support of the entry of chloride into cells was found in three of the cases reported here, W. H., W. L., and A. K. In the infant, Patient W. H., during therapy the chloride space expanded to a volume equivalent to approximately 50 per cent of the body weight which is unexpectedly large for a non-edematous subject even in childhood. A similar finding has been described during treatment of vomiting infants by Danowski and his associates (4), who were unable to attribute it to error in measurement of the chloride balance. In Patients W. L. and A. K. an inulin volume of distribution was determined at the end of replacement therapy. When this volume of distribution was taken as the final volume of extracellular fluid and, from it, and from the chloride balance, the initial volume of extracellular fluid was calculated, the latter was found to be less than 1 l. Since such a value is absurd, the conclusion is that either the measurement of the volume of distribution of inulin was in error, or that chloride entered cells. A definitive answer to this problem requires the accurate measurement of the extracellular fluid volume by some independent means, such as inulin, both before and after treatment, so that the intracellular transfer of chloride, if any, can be measured directly. Unfortunately, an opportunity to do this in the present study was not available. The possibility remains, therefore, that chloride entered the intracellular phase during treatment in these cases. But again because of the magnitude of the transfers involved, it seems most unlikely that this did occur to such an extent as to preclude the extrusion of sodium.

Certain conclusions can be drawn in regard to the relationship of these intracellular electrolyte abnormalities to the extracellular hypochloremia and alkalosis. The production of intracellular potassium deficiency and of probable sodium excess by prolonged absence of potassium intake, appeared to result in hypochloremia and elevation of the extracellular bicarbonate concentration. This suggests that under conditions of biological equilibrium or the steady state of adequate renal adjustment, as predicted by Darrow (1), these intracellular disturbances are associated with a diminished tubular reabsorption of chloride despite the presence of hypochloremia. Such a concept receives further support from an analysis of the renal response to chloride administration in some of these cases, as presented in Table IV. In three of the cases tabulated, the extracellular fluid volume was expanding to such an extent that the administered chloride was insufficient to raise the extracellular concentration even if the kidneys had been able to conserve it, *i.e.*, the kidneys excreted less chloride then the increment calculated as necessary to displace the bicarbonate to normal levels. In other words, in these cases, the kidneys had no chance to show conditioning by another factor and the data cannot be used to imply that the circumstances required a maintenance of hypochloremia and alkalosis in the presence of chloride administration. In the other four cases of alkalosis, three of whom had hypochloremia (N. D., W. H., A. J., and J. C.), during the initial days of study chloride was excreted by the kidney in amounts sufficient to have displaced the high bicarbonate had the chloride been conserved. Thus the failure to adjust the abnormal concentrations in the latter cases was not due to unavailability of chloride when neutral salts, NaCl and KCl, were given. Pitts, Ayer and Schiess (29) have found that, during the acute administration of sodium bicarbonate, as the tubular reabsorption of HCO_3 rises to a maximal rate, the reabsorption of chloride is correspondingly depressed; and as a result the total cation concentration in the extracellular fluid is undisturbed. The cases reported here are hardly comparable to these

experimental subjects of Pitts since ours represent prolonged states of equilibrium maintained in the presence of conditions other than acute loading of excess sodium bicarbonate. Furthermore, if the results of the acute i.v. administration of a single dose of sodium bicarbonate to a normal subject are observed over a period of many hours the initial rise in the rate of renal excretion of chloride is followed by a fall (30); as the elimination of the excess bicarbonate continues the kidneys again conserve chloride and the extracellular concentrations return to normal. In the last four cases shown in Table IV, therefore, at a time when chloride was available some other factors must have been conditioning the tubular reabsorption of this ion.

However, when chloride ion was administered rapidly and in large quantities as an acidifying salt, NH_4Cl, chloride was retained in sufficient amounts to raise its extracellular concentration to normal as the extracellular bicarbonate concentration was lowered. Simultaneously, the extracellular volume or chloride space expanded and sodium moved into it from the intracellular space. This occurred without any restoration of potassium to the cells. There are at least two interpretations of these transfers. One is that an excess of intracellular sodium may have made this adjustment possible. Another is that the kidney was unable to excrete at least part of the suddenly imposed load of extra anion with ammonia because of renal damage or because of the time lag inherent in the mechanism of ammonia formation. Under these circumstances, the transfer of cellular sodium (the only sodium available) was the only way to mitigate an otherwise abrupt change in acid-base equilibrium due to displacement of bicarbonate as CO_2 through the lungs. Such a sodium transfer requires some accounting in regard to the intracellular anion pattern; this problem is discussed below. But whichever of the above interpretations is accepted, of the response to NH_4Cl, it would appear that if there is an obligatory relationship between the extracellular concentrations of chloride and bicarbonate and intracellular cation compositions, the relationship pertains more directly to intracellular sodium than to potassium.

According to the Darrow hypothesis, the direct withdrawal of chloride in excess of sodium by way of gastric fluid, resulting in a metabolic alkalosis, should produce changes in intracellular cation composition even in the presence of an adequate intake of potassium. Unfortunately our data are incomplete on this point. Only in Patient T. W. were balance studies obtained under such conditions. Intracellular potassium was diminished because of the loss in the gastric washings, but intracellular sodium was not increased. This may be accounted for by the presence of moderately severe renal insufficiency which certainly interfered with any renal readjustments that might have been made. Prior to study, Patient H. H. had lost a large amount of gastric fluid by lavage. Studies during his replacement therapy showed a very large decrement of cellular sodium as his bicarbonate concentration fell, without much retention of potassium. Thus under some circumstances extracellular chloride deficit and alkalosis probably can be produced without necessarily affecting either the sodium or potassium content of the intracellular phase. This is in agreement with the experience of Evans and co-workers who found in both surgical patients and in dogs that alkalosis could occur without cellular potassium depletion and could be corrected without the administration of this ion (31–33).

Darrow has suggested that the primary factor in extracellular fluid which conditions the intracellular electrolyte pattern is the concentration of hydrogen ion rather than that of bicarbonate or chloride. In support of this thesis he has cited his experiments (34) in which rats on exposure to low atmospheric pressures developed sodium excess and a questionable potassium deficit in the cellular phase of skeletal muscle. Presumably an alkalosis developed due to primary CO_2 deficit with a *lowered* CO_2 content of extracellular fluid, although no such measurements were made. We have studied patients representing the converse of this condition, namely, those with respiratory acidosis due to the primary retention of CO_2 as a result of severe pulmonary disease. This condition leads to a secondary fall in chloride and rise in bicarbonate concentration as a response to the rise in level of carbonic acid. In a small series of such patients with marked elevations of serum bicarbonate concentration, but with *low blood pH*, the serum concentration of potassium was found to be uniformly within the normal range (35). This finding suggests, but does not prove, that no severe intracellular depletion of potassium coexisted

with the high bicarbonate concentration found in respiratory acidosis. These studies are not adequate controls to those reported in this paper since the uptake of potassium administered to such patients was not tested. But they do suggest that intracellular electrolyte composition is conditioned not by concentrations of extracellular bicarbonate but by concentrations of hydrogen ion or carbonic acid. The latter is suggested by data of Wallace and Hastings (36) who found by direct analysis of muscle that the bicarbonate content of intracellular fluid is very little affected by the bicarbonate of the extracellular phase (the cell is impermeable to HCO_3^-), the calculated intracellular pH being altered according to changes in CO_2 pressure alone.

Extensive differential net transfers of cations versus anions between the two phases of body fluids require consideration of their effect on the ionic electrical neutrality and osmotic pressure of the respective phases. Knowledge of the ways in which electrical neutrality of intracellular fluid is maintained under these circumstances awaits at least the complete description of net exchanges across the cell boundary of the cations, calcium and magnesium, in addition to sodium and potassium, and of the anion, phosphate, as well as chloride. Some evidence on this score has been supplied by Gardner, MacLachlan and Berman (37) who found in the skeletal muscle of potassium deficient alkalotic rats, that the intracellular bicarbonate was decreased and phosphate increased, and that calcium and magnesium, as well as sodium entered the cellular phase. However, in addition to external transfers, metabolic processes and variations in cellular pH must affect the degree of dissociation of the organic phosphate and proteinate buffer system within the cell. Alterations in cellular osmolarity, so produced, apparently share in conditioning movements of water (38). Therefore, any complete accounting of the maintenance of electrical neutrality and of osmotic equilibrium is impossible with the data at hand.

From the study of these cases it appears to the authors that a variety of disturbances in intracellular cation pattern may exist in the presence of metabolic alkalosis, produced in a variety of ways. These disturbances may consist of (*a*) a decrease in potassium and an increase in sodium, (*b*) an increase in sodium without change in potassium, and (*c*) a decrease in potassium without an increase in sodium. The first of these, *a*, is most common and is usually present when there has been over-activity of the adrenal cortex or when the intake of potassium has been very low for a prolonged period of time. Under the latter circumstance the kidneys continue to excrete potassium for a long enough period to produce a significant deficiency before renal conservation takes place. Likewise, prolonged loss of gastric fluid, usually associated with inadequate potassium intake, may become associated with a decrease in potassium and an increase in intracellular sodium. On the other hand abrupt and drastic disturbance of extracellular fluid composition, such as may occur with excessive vomiting or postoperative gastric lavage, may be associated with no intracellular changes or with other patterns of change such as *b* or *c* listed above. In such conditions are more likely to be found the renal changes described by Burnett, Burrows and Commons (12) leading to potassium wasting, and in such conditions is less likely to be found the steady state of biological equilibrium (Darrow's prerequisite to intracellular pattern, *a*). It would be hazardous, therefore, to predict the precise intracellular cation disturbances in any case of metabolic alkalosis without due consideration of the physiological and clinical circumstances of its origin.

SUMMARY AND CONCLUSIONS

By use of the balance technic the intracellular cation exchanges have been studied in ten cases of metabolic alkalosis during treatment.

In all but one of the cases potassium was retained in the intracellular phase. In seven of the ten cases the chloride balance was much more positive than that of sodium. On the assumption that chloride is extracellular, large amounts of sodium were calculated to have left the intracellular phase. In some patients the decrement of sodium was roughly equivalent to, and in others greatly exceeded, the increment of potassium.

The extracellular bicarbonate concentration fell to normal levels concurrently with the increase in potassium and decrease in sodium in the intracellular phase, in four cases. In three others when NH_4Cl was given, the bicarbonate concentration fell and sodium was extruded from the cells prior to any significant uptake of potassium by the latter.

In one of these cases no potassium was retained, and in one sodium entered cells as the alkalosis was corrected.

A variety of etiological factors appeared to be involved in this group of cases as follows: low potassium intake alone, two; low potassium intake plus gastrointestinal fluid loss, six; adrenocortical hyperfunction (ACTH therapy, carcinoma), two. No evidence of abnormal renal wasting of potassium was found and in two cases an extreme degree of conservation of the ion with U/P ratios of less than 1.0 was observed.

It is concluded from these data that although decrease in potassium and increase in sodium is a common intracellular cation disturbance in metabolic alkalosis, other patterns of disturbance may occur, especially when the onset has been abrupt and renal adjustment has not taken place. Where there is an equilibrium state between extracellular bicarbonate concentration and intracellular cation transfers, the former is more directly linked to those of sodium than to those of potassium.

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INTRACELLULAR CATION EXCHANGES IN ACIDOSIS DUE TO RENAL INSUFFICIENCY. EFFECTS OF ALKALI THERAPY [1]

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Disturbances in the exchanges of certain intracellular electrolytes which occur in patients with metabolic alkalosis, have been described in the previous paper (1). Clues to the direction and magnitude of these disturbances were obtained by calculating from balance measurements the alterations in body fluids produced by replacement therapy. In the present communication the results are presented of a similar study in patients with metabolic acidosis due to renal insufficiency.

Since this study involved the administration of hypertonic solutions of sodium bicarbonate or sodium lactate, it seemed advisable to reassess all of the factors, including those of intracellular transfers, which determine the quantitative effect of such replacement therapy on the total fluid and acid-base equilibrium of these patients. Previous studies have been made of the effects of alkalinizing sodium solutions. Palmer and Van Slyke (2) and later Hartmann and Senn (3) found that the rise in serum bicarbonate was such that the dose of alkali was apparently distributed through a volume equal to two-thirds of the body weight, a fraction which they believed represented the total body water. Since both bicarbonate ions (4, 5) and the major portion of sodium ions are not freely diffusible across the cell membrane, some explanation must be sought for the discrepancy between the value stated above and the now commonly accepted magnitude of extracellular fluid of 15 to 20 per cent of the body weight (6).

One of the factors which might explain this difference in the data of Palmer and Van Slyke and Hartmann and Senn was the fact that the dose of administered sodium salt was not corrected for that portion which was unabsorbed or excreted. Another factor was the failure to allow for the change in protein anion concentration in red cells and plasma. Since the amount of cation which is buffered by protein changes as pH is altered, Singer and Hastings (7) have indicated that the effect of administered acid or alkali is more directly measured by the change in "buffer base," which is equivalent to the change in bicarbonate plus protein anion, than by the change in bicarbonate alone. Such calculation applied to the data of Shock and Hastings (8) gives an apparent volume of distribution of orally administered sodium bicarbonate of about 45 per cent of the body weight, instead of the larger percentage obtained by Palmer and Van Slyke.

Since it seems unlikely that these large volumes of distribution of administered sodium bicarbonate or lactate can be explained by excretion or failure of absorption of sodium, intracellular transfers would appear to be involved. Darrow and coworkers (9) observed that in the muscles of rats rendered acidotic experimentally, the intracellular sodium was depleted. On the basis of this finding he has suggested (10) that uptake of sodium by cells is the explanation of the large doses of alkali required to combat clinical acidosis. Our study confirms this hypothesis and reveals that even when the errors of absorption and excretion are eliminated, and when the buffer base is measured, the quantitative results of alkali therapy in this type of acidosis can only be explained by extensive transfers of cations across the cell boundary.

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[2] Established Investigator of the American Heart Association.

[3] Fellow of the Department of Medicine.

EXPERIMENTAL MATERIAL AND METHODS [4]

Eight adult patients with metabolic acidosis due to renal insufficiency were studied. The diagnoses are listed in Table I. Four of these patients were in the polyuric phase of uremia and four were oliguric or anuric. The studies extended from two to ten days over the periods during which sodium bicarbonate was being administered by vein or by mouth. Balances were measured and electrolyte and water exchanges were calculated as in the previous paper (1).

In four of the patients during early periods of therapy detailed studies were made of changes in the acid-base equilibrium. This was done by the technic described by Shock and Hastings (11) which involves the micro-determination of pH and total CO_2 content of cutaneous blood. From these values and that of the hematocrit the tension of carbon dioxide (pCO_2) and the concentration of buffer base ([BB]) were calculated according to the nomogram of Singer and Hastings (7). These changes were then correlated with the data for the corresponding balance periods.

[4] The laboratory assistance of Mrs. Helen Lightman and Mrs. Claire Tissari is especially acknowledged.

Since in most of the cases large changes occurred in the urea nitrogen concentration of body fluids, some difficulty was involved in the calculation of transfers of intracellular potassium which were "in excess of nitrogen" or unrelated to catabolism and anabolism of cells. To make this calculation the external nitrogen balance was corrected by a value obtained by multiplying the change in blood urea nitrogen concentration by an assumed value for total body water. Obviously, where the change in blood urea nitrogen concentration is large and the external nitrogen balance is small, the derived value for "excess potassium" can be but a rough approximation. Such a calculation is useful, however, in indicating at least the direction and type of exchanges involved.

RESULTS

The results are presented in Tables I to III, and in Figures 1 and 2.

Sodium exchanges and acid-base equilibrium. During the administration of sodium bicarbonate, all of the patients were in a strongly positive sodium balance (Table I). The concentration of

TABLE I

External exchanges of water, electrolytes, and nitrogen

Patient	Age and sex	Diagnosis	Period	Average daily										Total period balance*			
				Intake					Urinary output								
				H_2O	Cl	Na	K	N	Vol.	Cl	Na	K	N	Cl	Na	K	N
			day of study	*ml.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*ml.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*
Group I: Polyuric patients																	
E. G.	59 F	Chr. pyelonephritis	0–4	2,870	70	221¶	82	10.7	741	28	40	14	2.5	+165	+725	+274	+32.8
C. P.†	19 M	Chr. glomerulonephr.	0–1	4,100	57	226	135	10.3	3,775	130	145	72	17.5	− 80	+ 75	+ 55	− 7.5
			1–2	3,200	60	226	135	10.7	2,285	69	82	55	9.1	− 10	+142	+ 80	+ 1.6
H. H.	70 F	Lower nephr. nephrosis	0–7	3,510	55	124	75	9.3	2,480	70	96	14	10.1	−111	+185	+429	− 6.0
A. T.	38 F	Tubular insufficiency	0–3	3,023	0	120	64	0	1,362	45	29	16	4.2	−141	+268	+143	−12.6
			3–10	3,747	99	108	117	4.3	1,802	88	93	42	8.7	+ 67	+ 86	+536	−30.2
Group II: Oliguric and anuric patients																	
G. D.‡	18 F	Dissem. lupus	0–2	1,170	23	300	1	—	140	5	1	1	—	− 2	+483	− 20	—
J. R.	36 M	Nephrosclerosis	0–1	1,120	36	251	2	0	140	2	1	5	0.4	+ 18	+244	− 6	− 0.5
			1–5	1,286	67	202	10	0.2	334	5	5	15	1.9	+237	+777	− 24	− 7.2
M. B.§	65 M	Lower nephr. nephrosis	0–6	1,800	27	111	0	0.5	105	7	12	2	0.8	−220	+359	− 41	− 9.3
I. C.‖	40 F	Lower nephr. nephrosis	0–8	1,505	24	118	0	0	5	—	—	—	—	− 42	+752	−135	− 9.1

* Balances corrected for small quantities of Cl and Na lost in serum drawn for analysis, and for electrolytes lost in gastrointestinal and pleural fluids where indicated.
† 200 ml. of vomitus included in balance.
‡ 600 ml. of pleural fluid included in balance.
§ Gastric drainage removed during last three days included in balance.
‖ Upper gastrointestinal drainage and electrolytes removed by resin enemata included in balance.
¶ Given by mouth.
Balance data are expressed per individual period rather than cumulatively.

TABLE II

Analyses of blood and serum, and changes in the distribution of sodium and potassium between the phases of body fluid

Patient	Time	Weight	Serum					Blood urea N	Extracellular volume†	Change in			
										Sodium		Potassium	
			CO_2	Cl	Na	K	H_2O*			Extracell.	Intracell.	Extracell.	Intracell.‡
	day of study	*kg.*	*mM. per l.*	*m.eq. per l.*	*m.eq. per l.*	*m.eq. per l.*	*gm. per l.*	*mg. per cent*	*l.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*
Group I: Polyuric patients													
E. G.	0	48.7	13.5	101	129	4.6	931	107	(8.0)				
	4	50.2	23.0	91	139	3.6		88	10.4	+420	+305	± 0	+178
C. P.	0	66.0	15.8	97	132	5.0	939	132	(10.0)				
	1	66.0	21.2	92	136	5.1		144	9.8	+ 15	+ 60	± 0	+ 86
	2	66.0	26.5	95	141	5.0		144	9.4	− 10	+152	− 3	+ 79
										+ 5	+212	− 3	+165
H. H.	0	—	11.7	99	126	2.5	942	112	(15.0)				
	7	—	21.6	103	139	2.5		104	13.5	− 15	+200	− 4	+428
A. T.	0	—	3.8	120	136	2.6	936	15	7.9				
	3	—	13.1	106	134	2.6	937	39	7.8	− 30	+298	− 1	+192
	10	43.6	25.6	99	134	4.1	934	20	(9.0)	+164	− 78	+17	+568
										+134	+220	+16	+760
Group II: Anuric patients													
G. D.	0	70.0	10.8	88	116	7.1	957	152	(15.0)				
	2	—	20.7	94	128	7.1		152	14.1	+ 65	+418	− 7	− 13§
J. R.	0	—	16.7	76	112	4.9	944	98	(16.0)				
	1	—	18.9	75	117	5.3	942	116	16.4	+125	+119	+ 9	+ 6
	5	—	27.0	75	131	6.1	940	148	19.2	+595	+182	+30	− 1
										+720	+301	+39	+ 5
M. B.	0	82.0	19.8	84	131	4.9	928	83	(17.0)				
	6	—	28.0	69	139	7.1	921	138	17.4	+210	+149	+41	+ 6
I. C.	0	—	17.6	93	135	5.3	931	116	(15.0)				
	8	—	21.6	76	143	5.4		224	17.8	+530	+222	+16	− 4

* Calculated from the concentration of serum protein.

† Calculated by changes in chloride balance forward or backward from an assumed extracellular volume (designated by parentheses).

‡ In excess of nitrogen.

§ Total intracellular K, not K in excess of N.

Time as designated by day indicates end of period at which time serum analyses were made, balances for the period determined, and exchanges calculated.

serum sodium rose in all (Table II). In all of the patients sodium entered the intracellular or "non-chloride" phase (Table II). This increment of intracellular sodium varied from 29 to 108 per cent of the amount of sodium retained, and in four of the eight patients was more than 50 per cent. In three of the eight patients little or none of the sodium was retained in the extracellular phase; consequently, in these patients, the increase in *concentration* of extracellular cation which was available for the bicarbonate and protein anions was the result primarily of decreases in extracellular water and chloride rather than of an increase in the *absolute amount* of extracellular sodium. Thus, whether or not there was an increase in the absolute amount of extracellular sodium, all of the

TABLE III

Acid-base data on cutaneous whole blood

Before and after initial periods of $NaHCO_3$ therapy

Patient	Time	Observed data			Derived data	
		Relative cell volume	pH	CO_2 content*	pCO_2†	Buffer base‡
	day of study§	*per cent cells*		*mM. per l.*	*mm. Hg*	*m.eq. per l.*
C. P.	0	19	7.24	13.2	31	32
	1	20	7.34	18.9	37	39
A. T.	0	41	7.00	4.5	19	21
	3	29	7.35	11.2	22	33
G. D.	0	20	7.34	10.6	21	31
	2	15	7.35	17.9	33	37
J. R.	0	26	7.23	12.7	31	32
	1	26	7.28	15.7	35	36

* Normal range is 20 to 24 mM. per l.
† Normal range is 35 to 48 mm. Hg.
‡ Normal range is 46 to 52 m. eq. per l.
§ Days of study correspond to those in Tables I and II.

patients became less acidotic: the pH, the pCO_2, and the concentration of buffer base of whole blood all returned toward normal levels (Table III).

Potassium exchanges. The serum concentration of potassium was within or below normal limits in the group of polyuric patients, and was elevated in oliguric patients (Table II). Since potassium was given to the former group alone, only this group exhibited a positive potassium balance (Table I). In the anuric group the negative potassium balance reached large proportions only in Patient I. C. to whom cation exchange resin ene-

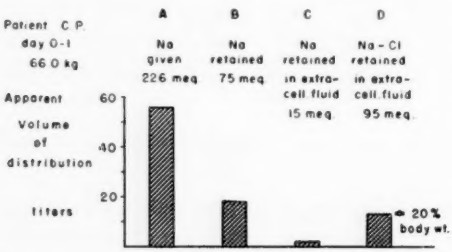

FIG. 1. APPARENT VOLUME OF DISTRIBUTION OF VARIOUS INCREMENTS OF ADMINISTERED SODIUM BICARBONATE

Volumes of distribution are calculated by dividing the increment of sodium indicated above the columns by the change in concentration of serum sodium (Columns A and B) or by the change in concentration of buffer base (Columns C and D).

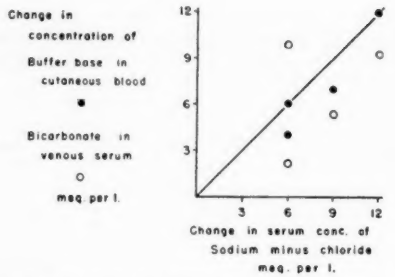

FIG. 2. CHANGES IN SERUM CONCENTRATION OF SODIUM MINUS CHLORIDE COMPARED WITH THOSE IN BUFFER BASE OF CUTANEOUS WHOLE BLOOD AND WITH THOSE IN BICARBONATE OF VENOUS SERUM, INDUCED BY THE ADMINISTRATION OF SODIUM BICARBONATE

mata were administered (12). In the anuric patients no significant exchanges of "excess" potassium took place, but in the polyuric patients potassium in excess of nitrogen entered the intracellular phase (Table II). The largest uptake of "excess" potassium was in Patient A. T. and was equivalent to 17.4 m. eq. per kilogram of body weight.

DISCUSSION

These data support the evidence of Darrow and his co-workers that intracellular sodium may be depleted in at least some types of metabolic acidosis. Certainly administered sodium ion was taken up by the "non-chloride" space in our patients, and to a degree that was surprising. The observation that this portion of sodium varied from one-third to all of the ion retained emphasizes the necessity of taking this phenomenon into consideration when estimating the therapeutic dose of sodium ion under these circumstances. As Darrow predicted, this transfer of sodium into the intracellular phase readily explains the large doses of alkali which on an empirical basis have been found necessary to successfully combat an acidosis in terms of raising a depressed concentration of extracellular bicarbonate or of buffer base.

Thus it is patently absurd to consider in clinical practice that all of the sodium bicarbonate administered, or even retained, enters into the correction of the extracellular electrolyte abnormality which defines the metabolic acidosis. This is well illustrated in Figure 1, in which are shown the volumes of distribution of various increments of so-

dium bicarbonate administered in one day to Patient C. P. Only part of the sodium given was retained and of this some entered the intracellular phase. Furthermore, the increment of sodium in the extracellular phase had to be corrected by subtracting the balance of chloride since a fall in chloride as well as a rise in sodium concentration increases the base equivalent of bicarbonate and protein in extracellular fluid. When this corrected increment of extracellular sodium minus chloride was divided by the rise in buffer base concentration, a volume of distribution was obtained which approximates a reasonable value for the volume of the extracellular fluid. This is in sharp contrast to the larger values obtained from the data of the previous workers (2, 3) mentioned in the introduction above, who failed to take all of these factors into consideration.

These studies also indicate that measurements of changes in concentration of the whole blood buffer base more accurately gauge the effect of therapy on the acid-base balance than do those of bicarbonate or total carbon dioxide. In Figure 2 the changes in serum concentration of sodium minus chloride, induced therapeutically in four of the patients, are compared with the changes in concentration of whole blood buffer base and venous serum carbon dioxide. The correlation is better with the buffer base than with the bicarbonate because the former takes into account the alterations in anionic proteinate due to pH shifts. Failures to show perfect correlation are accounted for by changes in concentration of other undetermined anions such as phosphates, sulfates, and organic acids, by analytical error, and by the effect of stasis on the carbon dioxide content of venous blood. The calculation of buffer base from pH and CO_2 content of cutaneous blood obviates two of these errors and appears to represent a real improvement in accuracy over the venous serum carbon dioxide concentration in following metabolic disturbances of acid-base balance.

Our data emphasize the extreme difficulty of calculating accurately the amount of sodium required to correct a metabolic acidosis. Even if one can estimate the amount of administered sodium that is likely to be excreted, the increment that will enter the intracellular phase is at present quite unpredictable. Because of these uncertainties in the sodium exchanges, alkali therapy of acidosis can be at best but a matter of intelligent trial and error.

The direction of the exchanges of potassium was not unexpected. Retention of potassium with elevation of the extracellular concentration in renal insufficiency with oliguria and anuria has been described by many workers (13–15). In such cases it is apparent that the potassium intake should be rigidly restricted since viable cells do not readily take up the ion which is in excess in the extracellular phase (15). Potassium deficiency in renal insufficiency is less common but does occur (16). Such a deficiency undoubtedly existed in two of the polyuric patients reported here (H. H. and A. T.) since potassium was taken up by the cellular phase in excess of nitrogen in amounts quite comparable with those retained in other types of potassium deficiency (17). The mechanism of such a deficiency is not clarified by our present studies. No evidence is presented that tubular secretion occurred in normal or excessive amounts or that tubular reabsorption was diminished. Both of these mechanisms have been implicated in severe renal disease (18, 19). A subsequent study of Patient A. T., with the unusual renal lesion of "tubular insufficiency without glomerular insufficiency" (as first described by Albright and co-workers [20]), confirmed the fact that under the stress of the need to excrete excessive amounts of anions, potassium excretion was increased (21). It is probable, however, that in most cases of renal insufficiency in which potassium deficiency occurs, it does so at least in part because of a diminished intake of the ion in food as the result of anorexia and vomiting.

This study re-emphasizes the fact that exchanges of intracellular sodium are not necessarily reciprocal to those of potassium (22). In the polyuric patients sodium ion entered the cellular phase simultaneously with potassium; in the anuric patients a similar transfer of sodium took place without any exchange of "excess" potassium. Apparently, in metabolic acidosis, as in alkalosis (1), the disturbance in acid-base equilibrium is more closely correlated with changes in the sodium content than with changes in the potassium content of the intracellular phase. The patterns of such intracellular disturbances are undoubtedly conditioned by many factors including those of time, renal function, nutrition, and exchanges of water

and of other ions. As pointed out in the preceding paper (1), any assessment of the relation of these cation transfers to maintenance of ionic electrical neutrality and to osmotic equilibrium within the cells, requires a great deal more data than is available at present. Precise knowledge of these disturbances will only come when these other factors have been more adequately studied and their relationship defined to the metabolic processes which maintain the differential distribution of electrolytes across the cell boundary.

SUMMARY AND CONCLUSIONS

The exchanges of intracellular sodium and potassium were calculated by the balance technic in eight patients with acidosis due to severe renal insufficiency, during the administration of sodium bicarbonate. On four of the patients acid-base data were obtained.

Sodium entered the intracellular phase in all of the patients in amounts ranging from 29 to 108 per cent of the sodium retained.

Potassium entered the intracellular phase in the four polyuric patients to whom it was given. In the anuric patients to whom no potassium was given because of an elevated extracellular concentration of the ion, no significant exchange occurred of intracellular potassium in excess of nitrogen.

The metabolic acidoses as measured by the buffer base or bicarbonate were corrected in whole or in part in all of the patients.

It is concluded *(1)* that measurements of the whole blood buffer base give a better index than the CO_2 content of changes in this type of acid-base disturbance, *(2)* that shift of sodium into cells, as well as excretion of sodium and balance of chloride, determine the effect of administered alkali on the acidosis, and *(3)* that because of the unpredictability of these factors the therapeutic dosage can only be roughly estimated ahead of time.

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THE LIPIDS OF SERUM IN PREGNANCY[1]

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It has been repeatedly demonstrated that the concentration of cholesterol in the serum of women increases during pregnancy (1–8). Fatty acids (1, 6, 7) and phospholipids (4, 6, 7) also rise somewhat. There is not such general agreement concerning the time relations of the elevations nor the relative changes of the various lipid components. Bloor and Knudson (9) claimed that the increment of cholesterol is composed chiefly of esters; Gardner and Gainsborough (5) found that it consisted entirely of free cholesterol; while, according to Boyd (7), the partition of cholesterol is unaltered. These disagreements can probably be attributed chiefly to the different analytical techniques employed.

In a study of the effect of pregnancy on the precipitable iodine of the serum, lipids were also measured in a number of cases in order to define more precisely the course of the lipids during pregnancy and also to evaluate the significance of the changes of SPI during this process.

MATERIAL AND METHODS

The patients studied were under the supervision of members of the metabolic division of the Department of Internal Medicine. The majority were delivered in the New Haven Hospital. Blood was taken in the morning. The patients were either fasting or else had a meal containing carbohydrate, but no appreciable amounts of fat or protein (fruit juice, black coffee with sugar, toast without butter, with a little jam or jelly if desired). SPI and serum lipids were measured by methods previously described (10).

RESULTS

Altogether 145 observations were made on 34 normal persons and 19 observations on five patients with hyperthyroidism under treatment with propylthiouracil and urea. SPI and all lipid fractions (total and free cholesterol, fatty acids and lipid phosphorus) of 31 normal subjects were measured on 114 occasions. In one instance SPI was omitted and in a number of others fractionation of lipids was incomplete.

The data from the normal subjects are presented in Table I. Although the interrelationships of the lipid fractions are not entirely unaffected by pregnancy, in general all fractions are altered synchronously and in the same direction. In every case in which repeated observations were made the lipids rise as pregnancy advances. The rise, which is extremely variable in degree, is seldom evident before 12 or more weeks have elapsed. Once it has begun, the lipids appear to increase progressively until term. After delivery they decline gradually, presumably to prepregnant concentration. The word "presumably" is used because, for obvious reasons, actual prepregnancy values are not known. There is reason to suspect that the concentrations observed in early pregnancy do not represent normal nonpregnant values. With a few exceptions, until the lapse of 12 weeks they tend to lie somewhat below the accepted average for a group of normal individuals. In a number of subjects who had more than one observation in the early weeks, cholesterol declined distinctly at this time: MMcM, C46864, MWa. The cholesterol of PG after 10 weeks of pregnancy was 212 mg. per cent; but 26 weeks after delivery it was 275 mg. per cent. Of course it may not have returned to its normal level in this interval, but it had remained at the same concentration for 15 weeks, definitely lower than it had been in the latter part of pregnancy. The first case, 43773, is particularly significant because she was studied in two pregnancies. Before the first pregnancy only lipid phosphorus was measured. Values of 9.9 and 10.2 mg. per cent were found on two occasions. It had reached practically the same value, 10.5 mg. per cent, 32 weeks after delivery. In the early weeks of both pregnancies, however, it fell to between 8 and 9 mg. per cent. As evidence that her condition did not differ appreciably on the two occasions, her lipid patterns at 17 and 18 weeks of

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TABLE I

Lipids in normal pregnancy

Case no.	Age	Weeks*	SPI	Cholesterol			Fatty acid	Lipid P	Neutral fat fatty acid	Lipid P − 3.7 / Free chol.
				Total	Free	Free / Total				
			γ %	mg. %	mg. %		mEq/L	mg. %	mEq/L	
43773	29	a 5						9.9		
		a 2						10.2		
		7						8.3		
		17		185	51	0.27	12.3	9.6	3.2	0.116
		30	7.1	215	55	0.25	14.7	11.0	4.1	0.133
		33	5.8	211	64	0.30	14.8	11.3	4.4	0.119
		39	6.2	222	53	0.24	15.6	12.0	4.2	0.157
		p 2/7	5.9	221	45	0.28	14.1	11.9	2.6	0.182
		p 12/7	4.4	217	69	0.32	12.2	11.1	2.0	0.107
		p 8	4.1	225	58	0.26	12.2	8.6	2.9	0.102
		p 32	5.1	213	63	0.30	13.2	10.5	3.2	0.108
		9	4.5	160	42	0.26	10.7	8.8	2.5	0.121
		13	4.9	165	48	0.29	9.9	8.6	1.9	0.102
		18	5.8	187	48	0.26	12.5	9.9	3.2	0.129
C41916	25	12	7.9		48		11.9	9.3		
		15	4.0	223	50	0.22	13.6			
		16	6.2	234	60	0.26	12.8	10.1	2.4	0.107
		20	6.6	243	70	0.29	17.7	10.5	7.1	0.099
		23	7.0	263	77	0.29	20.7	11.6	9.2	0.102
		27	7.3	316	94	0.30	23.4	14.3	9.6	0.113
		31	7.0	342	100	0.29	23.9	14.3	9.3	0.106
		33	6.9	332	90	0.27	22.5	13.3	8.5	0.107
		35	8.0	355			25.1	14.8		
		p 2/7	8.9	333	106	0.32	27.9	14.5	15.8	0.102
		p 6	5.7	242	75	0.31	12.5	10.3	2.4	0.089
PG	28	10	8.0	212	53	0.25	11.8	10.4	1.7	0.127
		14	9.5	273	75	0.27	17.3	13.0	4.7	0.124
		19	9.2	272	81	0.30	16.9	12.0	4.9	0.102
		23	10.0	340	99	0.29	20.5	14.2	6.0	0.106
		27	8.5	355	83	0.23	20.4	14.3	5.0	0.127
		33	9.0	379	111	0.29	22.3	14.9	6.6	0.101
		37	8.0	326	96	0.29	21.7	14.3	7.4	0.111
		p 5/7	7.9	349	84	0.24	16.5	13.2	1.8	0.113
		p 4	6.5	297	76	0.26	15.3	12.4	2.4	0.114
		p 11	5.0	271	75	0.28	16.3	12.7	3.8	0.120
		p 26	7.0	275	76	0.28	14.7			
DM	28	9	6.5	157	40	0.26	10.5			
		13	7.1	156			10.6			
		22	7.1	185	52	0.28	12.8	9.3	4.0	0.107
		27	7.3	179	49	0.28	14.4	8.9	5.8	0.106
		32	8.4	178			17.9			
		p 3/7	7.4	181	57	0.31	14.8	9.4	1.6	0.100
		p 6	4.7	146				9.7		
MMcM	20	4	4.9	172	47	0.27	11.6	8.8	3.3	0.109
		8	4.9	132	29	(0.22)†	8.3	6.9	1.6	0.114
		13	6.5	149	37	0.25	8.3	7.9	0.8	0.113
		20	8.9	156			11.9	9.1		
		23	7.0	193	45	0.23	16.1	9.5	6.8	0.129
		27	5.4	197	56	0.28	18.0	9.8	8.6	0.109
		p 6/7	9.4	233	64	0.27	13.7	10.1	3.4	0.100
		p 3	6.4	201	58	0.29	11.9	9.8	1.5	0.105
		p 8	5.5	222			13.1	8.0		
C46864	20	7	5.5	176	45	0.26	10.5	9.0	1.9	0.118
		8	6.9	173	46	0.27	9.0	8.6	0.7	0.107
		9	6.3	160	44	0.27	10.1	7.9	2.5	0.095
		12	6.9	166	51	0.31	10.3	7.7	2.8	0.075
		16	8.7	183	50	0.28	14.0	8.9	5.4	0.104
		22	9.8	219	63	0.29	16.3	11.9	5.3	0.130
		26	9.4	227	65	0.29	14.8	10.7	4.4	0.108
		31	8.4	246	72	0.29	16.4	11.3	5.3	0.106
		36	7.3	270	78	0.29	18.6	11.8	6.8	0.104
		38	8.9	252	71	0.28	16.4	11.9	4.8	0.115
		40‡	8.6	265	73	0.28	20.7	13.8	7.7	0.138
		p 4/7	11.2	277	80	0.29	18.2	14.0	4.8	0.133
		p 8	5.1	211	52	0.25	11.6	10.4	0.2	0.129

TABLE I—*Continued*

Case no.	Age	Weeks*	SPI	Cholesterol			Fatty acid	Lipid P	Neutral fat fatty acid	Lipid P − 3.7 / Free chol.
				Total	Free	Free / Total				
			γ %	mg. %	mg. %		mEq/L	mg. %	mEq/L	
B85841		12	6.7	183	54	0.29	12.0	9.9	3.0	0.114
		17	7.8	190	49	0.26	13.2	10.4	3.5	0.136
		20	6.9	212	63	0.30	13.1			
		24	7.4	239	70	0.29	15.7	11.8	2.7	0.116
		28	7.1	242	74	0.31	18.2	11.0	7.4	0.099
		32	7.1	242	80	0.33	19.5	12.7	7.9	0.112
		35	9.5	243	74	0.30	19.3	12.5	7.6	0.118
		36	7.7	260	86	0.33	19.8			
		p 1	8.6	244	76	0.31	16.4	11.5	5.3	0.102
		p 6	4.8	235	65	0.28	15.8	10.7	5.2	0.108
B78510‖	29	16	7.3	244	66	0.27	14.6	12.3	2.9	0.130
		21	6.4	238	89	0.37	18.5	12.4	7.4	0.098
		25	6.3	333	113	0.33	21.9	16.0	7.0	0.109
		31	7.4	350	144	0.41	23.0	16.4	8.1	0.088
		35	6.0	346	111	0.32	22.9	14.3	8.5	0.096
		39	8.8	423	111	0.26	24.9	15.0	8.1	0.102
		p 2/7	9.4	385	112	0.29	22.4	15.0	6.6	0.101
		p 11/7	7.3	385	109	0.28	18.4	14.9	2.5	0.103
		p 5	5.0	354	108	0.30	18.2	12.6	4.5	0.082
		p 12	4.5	292	80	0.27	14.8	11.0	2.9	0.091
		p 21	5.4	261	71	0.27	13.9	10.9	2.7	0.101
C52217	32	5	8.0	160	49	0.31	10.0	7.6	0.1	0.080
HC		12	8.3	168	45	0.27	9.3	9.8	0.4	0.135
		24	7.3	215	56	0.26	12.7	11.1	2.2	0.132
MD	22	8	8.6	176			10.7			
		p 2/7	6.7	264	82	0.31	18.2	13.5	6.2	0.120
B53121	37	6	4.6	205	64	0.31	12.6			
		7	4.4	230						
		32§	8.0	263	76	0.29	25.7	13.1	13.2	0.124
VD	34	36	8.4	367			29.8			
CF	23	11	6.9	225	68	0.30	14.8	12.9	3.2	0.135
C45201	22	14	7.6	213	60	0.28	13.5	10.6	3.4	0.115
C13678	26	3	4.0	108			6.4			
AH		10	7.6	197	62	0.32	11.1	8.8	2.5	0.082
C42461	23	5	6.0	143	39	0.27	8.1	7.8	0.9	0.106
		7	6.6	141	38	0.27	8.2	5.8	2.4	0.068
		13	9.0	141	31	0.22	7.4	6.1	1.0	0.077
		26	9.0	223	58	0.26	12.8	9.2	3.2	0.095
C15296	27	34	4.8	329	94	0.29	24.6	15.5	9.5	0.125
		40	4.8	266			31.5			
GJ	28	20	7.8	235	78	0.33	19.4	15.4	6.4	0.150
PJ		14	6.4	161	51	0.32				
B95180	29	32	6.5	252	79	0.31	19.2	13.8	6.7	0.128
MMcN		32	6.3	292	79	0.27	19.9	13.8	6.4	0.128
		37	6.6	296						
SO		8	4.3	201	59	0.29	12.9			
C41546	21	8	6.2	201	66	0.33	10.7	8.5	2.3	0.073
B769	29	12	7.0	225	60	0.27	11.4	9.9	1.4	0.105
MWa	28	7	7.6	146	38	0.26	9.6	6.3	3.1	0.068
		11	7.7	130	38	0.29	9.5	7.4	2.8	0.074

* Weeks after last menstrual period; a represents weeks before pregnancy; p represents weeks after delivery.

† Parentheses indicate that duplicate values for total or free cholesterol did not check with sufficient accuracy; therefore the ratios are not precise.

‡ One day before delivery.

§ One day before premature delivery. For details of case see (12).

‖ Received 0.02 gm. of stilbestrol daily from the end of the 16th week until delivery.

the two pregnancies respectively were almost identical. In the second pregnancy the early reduction of lipids involves all fractions proportionally. All these considerations together suggest that the lipids diminish in the early part of pregnancy. If this is the case the data tend to exaggerate the extent of the rise of lipids during pregnancy. This could also be inferred from the fact that the highest values observed in this series seldom exceed the normal limits. In only four subjects were concentrations of cholesterol greater than 300 mg. per cent encountered. The rise, nevertheless, appears to be authentic. The general range of lipids in the later part of pregnancy is distinctly above the expectation for a normal group of nonpregnant women. In addition, with the exception of 43773 and MMcM, who was followed for only eight weeks *post partum*, in those who were followed after delivery, lipids declined. The rises in these two cases were comparatively small. The rate of decline of the lipids is highly variable. There are not sufficient observations to define its course precisely. It apparently does not usually begin until a week or two have elapsed and may not be complete after as long an interval as 12 weeks.

In Table II are a few subjects who received thyroid although they had no evidences of hypothy-

TABLE II

Serum lipids of patients who, although they had no disorder of thyroid function, received thyroid

Case no.	Age	Weeks*	SPI	Cholesterol			Fatty acid	Lipid P	Neutral fat fatty acid	Lipid P − 3.7 / Free chol.	Thyroid†	
				Total	Free	Free / Total						
			γ %	mg. %	mg. %		mEq/L	mg. %	mEq/L		gms.	wks.
HL	42	26	7.6	236	65	0.28	17.3	11.8	6.1	0.125		
		31	5.5	235	63	0.27					0.12	3
		32	4.8	183	72	(0.39) ‡					0.12	4
GM	31	4	5.2	167	47	0.28	10.2					
		12	5.9	160	40	0.25	10.4	9.3	1.9	0.140	0.12	8
HP	35	7	6.3	160	83	0.52	9.5	9.7	1.9	0.072		
		9	9.9	188	49	0.26	12.2	10.6	2.5	0.142	0.18	2
ES	36	7	3.9	188	52	0.28	10.9	10.6	1.3	0.133		
		11	5.8	187	49	0.26	10.0				0.12	4
		15	7.4	169	47	0.28	11.0	9.3	2.4	0.119	0.24	3
		27	5.3	218	62	0.28	14.4	11.6	3.6	0.127	0.24	15
		35	5.4	255	71	0.28	16.5	11.8	4.9	0.114	0.24	23
A63436	35	5	3.5	194	47	0.24	13.6	10.1	3.9	0.136	0.24	1
		6	5.9	172	49	0.28	12.2	9.6	3.4	0.120	0.30	2
		12	8.5	169	35	0.21	11.4	10.1	2.0	0.183	0.30	8
		16	7.9	253	70	0.28	17.4	12.5	5.3	0.127	0.30	12
		18	8.1	221	62	0.28	17.2	11.8	6.3	0.130	0.42	2
		21	8.6	252	73	0.28	18.8	12.9	6.7	0.126	0.42	5
		23§	8.4	241	70	0.29	14.2	11.8	3.0	0.115	0.42	7
MW	22	1	4.2	236	60	0.25	14.4	10.0	3.5	0.128		
		5	5.5	202	56	0.28	11.6	9.0	2.6	0.095		
		9	7.2	165	49	0.30	8.8	8.0	1.2	0.088	0.18	3
		11	7.3	174			9.8				0.18	5
		15	7.8	190	56	0.29	12.0	9.1	3.2	0.096	0.18	9
		20	9.4	228			16.4				0.18	14
		25	7.7	252	73	0.29	17.0	11.9	5.5	0.111	0.18	19
		p 2/7	9.7	259	77	0.30	18.2				0	2/7
		p 15	4.1	233	70	0.30	12.3	10.5	2.3	0.090	0	15
TK	30	18	6.2	279	77	0.39	16.3	12.8	3.7	0.118		
		23	6.8	246	73	0.34	19.7	13.1	7.3	0.129	0.24	4

* Weeks after last menstrual period; p represents weeks after delivery.

† The number of weeks that the stated dose of thyroid had been taken before the determination.

‡ Parentheses indicate that duplicate values for total cholesterol did not check with sufficient accuracy; therefore the ratios are not precise.

§ On the day of miscarriage. For details of case see (12).

roidism. The general course of the lipids in these cases is not distinguished from that of the women who did not receive thyroid. This bears out the impression, expressed earlier (11, 12) that the pregnant woman tolerates or utilizes thyroid with facility. The lipids may be temporarily depressed after institution of thyroid therapy or an increase of the dose, but rise again shortly. This is evident in ES, A63436 and MW. Observations were not continued long enough in HL and TK to ascertain whether the drops were transitory or not.

Free cholesterol followed the course of total cholesterol quite closely. The ratio, free : total cholesterol, in the serum of normal persons and patients with disorders of the thyroid (10) does not depart from the narrow limits, 0.24 to 0.32. In 138 observations in this study the ratio exceeded 0.32 only 11 times and 0.33 only seven times. One of these, in HL (Table II), must be discounted because the analysis was not satisfactory. In TK (Table II) the chloresterol ratio was 0.39 after 18 weeks, 0.34 after 23 weeks. No disorder was found to explain these abnormal ratios. The pregnancy proceeded to a normal delivery. In HP (Table II) at the end of seven weeks the ratio was 0.52, but two weeks later it was only 0.26. A ratio of 0.34 was found in 98175 (Table III) two days after delivery. Ratios of 0.37, 0.33 and 0.41 were found in B78510 after 21, 25 and 31 weeks, respectively. This subject had normal ratios after 16 and 31 weeks and thereafter on seven different occasions, the last 21 weeks after delivery. This is the woman whose total cholesterol rose highest during pregnancy, reaching 423 mg. per cent at the end of 39 weeks. There was, however, no correlation between total cholesterol and the cholesterol ratio. The latter had fallen to 0.26 when the former reached its peak. The subject had had infectious hepatitis three years earlier, but had no demonstrable residua of this disease. She received stilbestrol throughout her pregnancy, being the only subject thus treated. However, the ratio returned to normal while she was still receiving stilbestrol, while the lipids remained high after it was discontinued. The ratio was less than 0.24 in

TABLE III

Serum lipids of patients with disorders of the thyroid

Case no.	Age	Weeks*	SPI	BEI	Cholesterol			Fatty acid	Lipid P	Neutral fat fatty acid	Lipid P − 3.7 / Free chol.	Lugol's solution	Propylthiouracil	
					Total	Free	Free / Total							
			γ %	γ %	mg. %	mg. %		mEq/L	mg. %	mEq/L		gtts.	mg.	wks.†
C24644	34	6	9.6		182	47	0.26	10.6	8.3	2.2	0.098	12	100	25
		14	11.3		253	74	0.29	16.3	11.9	4.8	0.111	12	75	7
		20	9.1		252	77	0.31	16.4	11.9			12	75	13
		30	8.9	7.2	276	86	0.31	20.2	13.9	7.2	0.119	12	75	23
		p 1/7	9.1	7.6										
		p 5	7.7	4.7	243			19.5	13.1			12	100	5
98175	32	10	9.4	6.9	257	73	0.28	16.8	10.4	6.0	0.072	15	450	6
		14	14.9	13.7	242	75	0.31	18.0	11.7	6.9	0.107	15	450	10
		21	8.6	6.4	342	94	0.28	24.6	15.3	9.3	0.099	15	500	7
		24		14.0	278	78	0.28	25.3				15	300	3
		26	14.9	12.9	318			25.2	14.6			15	300	5
		p 2/7		16.4	305	105	0.34	22.7	14.9	8.9	0.107	15	150	8
A25444	26	18	11.2	6.7	200	59	0.29	16.2	13.4	4.8	0.164	7	300	8
		23	8.6	6.1	349	73	0.21	19.1	14.0	3.8	0.141	7	300	13
		28		11.8	159	49	0.31	12.5				7	150	5
B94697	36	37	8.7		186	54	0.29	19.9	11.5	10.8	0.144	10	50‡	16
		40	11.3		189	59	0.31	20.2	10.9	10.5	0.122	10	25‡	3
		p 2/7	10.5		173			17.4				10	25‡	3
		p 2	11.1	6.8	270	72	0.27	21.1	11.6	9.2	0.110	10	25‡	5
C19922	36	19	9.2		179	48	0.27	15.2				14	300	14

* Weeks after last menstrual period; p represents weeks after delivery.
† Number of weeks patients had been taking propylthiouracil.
‡ Thiourea instead of propylthiouracil.

three subjects. One of these can be attributed to analytical error. No cause for abnormally low ratios has been discovered. In no case could variations of the ratio, within or outside of the normal range, be related to the stage of pregnancy or to other characteristics of the lipids. The deviations from the normal range must, therefore, be attributed to adventitious disorders, the nature of which was not discovered.

In normal persons the relationship of phospholipid to cholesterol is most precisely defined by the equation, $\frac{\text{mg. per cent of lipid P} - 3.7}{\text{mg. per cent of free cholesterol}} = 0.106$ (13). In the present study the average ratio was 0.113, varying from 0.067 to 0.182. No significance can be attached to the slightly elevated average ratio or the variability of the ratio. These features could not be correlated with the course of pregnancy, other lipid fractions or the cholesterol ratio.

The most distinctive feature of the lipid pattern is an increase of the concentration of neutral fat as pregnancy progresses. Although this is usually moderate and varies from case to case, it is unmistakable and consistent in all subjects and therefore significant. Neutral fat and cholesterol appear to be roughly correlated; but the correlation probably derives only from the fact that both rise as pregnancy progresses. Neither in individual cases nor in the series as a whole do the two functions vary proportionally. Neutral fat appears to fall earlier than cholesterol does after delivery.

Both SPI and cholesterol rise as pregnancy advances. There is no reason except this association to connect the two phenomena. In most other circumstances cholesterol tends to diminish as SPI rises.

In Table III are five pregnant women with hyperthyroidism who were treated with this drug and Lugol's solution of iodine. Although the lipids are more variable in these patients than they are in normal women they have the same tendency to rise as pregnancy advances and to decline after delivery. SPI in these cases does not mirror precisely the degree of thyroid activity for reasons pointed out elsewhere. SPI may rise further in hyperthyroid than in normal persons during pregnancy (12). In addition administration of iodine causes spurious increases of SPI (14). Butanol-extractable iodine (BEI), figures for which are given in several instances, is a better measure of thyroid activity in these cases. In A25444 and 98175 the lipids fell when signs of frank hyperthyroidism appeared with BEI's above 10 gamma per cent. In the latter case, they rose again in spite of the continued hyperthyroidism.

DISCUSSION

There is little to add to this description. The causes of these variations in the concentrations and interrelationships during pregnancy will remain conjectural until the general metabolism of pregnancy has been further elucidated. The early decline of lipids, if it is verified, may be related to vomiting of pregnancy, but this was not conspicuous in 43773 (Table I), whose lipids fell with the onset of both pregnancies. It has been shown that certain estrogenic materials increase the lipids of the serum and livers of rats and guinea pigs (15–17). Increases of neutral fat are reported to occur either after meals or when fat is being mobilized rapidly from the depots to the liver for formation of ketone bodies. It is intriguing to speculate about the possible connection of this phenomenon in pregnancy with the recognized tendency of the pregnant woman to spend carbohydrate prodigally and her unusual susceptibility to ketosis. Variability of the decline of lipids after delivery may be connected with lactation; but no such connection could be discerned in this small series of subjects. If the hyperlipemia were related to the great discharge of estrogenic materials that characterizes pregnancy such a delay should not be expected.

The association of hyperlipemia with high SPI might be interpreted as evidence of relative hypothyroidism. It is conceivable that the demands of the tissues for the thyroid hormone and utilization of the hormone are augmented during pregnancy and that the increased discharge of the hormone manifested in the elevated SPI is not sufficient to meet these demands. This is an unlikely explanation for two reasons. When the hyperthyroid patient, 98175, lapsed into overt hyperthyroidism, although the lipids fell they still remained far higher than they would have been in a hyperthyroid nonpregnant woman. Moreover in the hyperlipemia of hypothyroidism neutral fat does not rise (10).

SUMMARY AND CONCLUSIONS

The lipids and precipitable iodine (SPI) of the serum of 34 normal women and five women with medically treated hyperthyroidism have been measured at intervals during pregnancy and after delivery. There is evidence to suggest that the lipids may decline slightly in the early weeks of pregnacy. After about the 12th week, however, they rise progressively until delivery, thereafter declining at a variable rate. In this rise total and free cholesterol and phospholipids participate proportionally, maintaining their normal relations to one another. Neutral fat, however, rises proportionally far more than the other lipid fractions and declines more rapidly than the others after delivery.

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THE BIOLOGICAL ACTIVITY OF A FLAVONOID (VITAMIN "P") COMPOUND [1, 2]

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The elaboration of a practical and reliable biological test for the estimation of vitamin "P" activity is the most urgent and important problem with which every investigator in this field is confronted. The complexity of the physiological activity of flavonoid compounds was responsible for the obstacles in finding a satisfactory solution of this problem. The attempts to induce experimental avitaminosis "P" gave contradictory observations making this test unreliable for any practical use (1–8). A method was suggested for the estimation of vitamin "P" activity by using the capillary bed in the rat-mesoappendix, with a compound topically applied. It was found, however, that most of the flavonoid compounds, except catechins, did not produce any vasoconstrictor effect (9, 10). Considerable work was done with the determination of capillary permeability by using chloroform wheals applied to the skin and simultaneous injections of trypan blue. This test pertaining rather to permeability changes than to increased capillary fragility has demonstrated a great variation in the response of experimental animals to the same flavonoid compound (11, 12). More recently it was suggested that radiation injury to the capillary wall might serve as a basis for the estimation of vitamin "P" activity. Purpura, for which ascorbic acid has proved ineffective, has been a constant feature of radiation illness. It has been shown that some flavonoid compounds exert protective action against radiation injury to the capillary system (13–18).

In our present report which covers both experimental and clinical data particular attention was paid to the determination of vitamin "P" activity in the condition where increased capillary fragility rather than permeability changes were evidenced.

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MATERIAL

A flavonoid compound isolated from citrus fruit was used for our experimental and clinical work. It contained four identified factors; namely, hesperidin-chalcone, glucose-hesperidin chalcone, eriodictin and quercitrin-like substance, which seemed to form a complex molecule as U.V. readings have shown. It was a yellow powder slightly hygroscopic, with impurity of 0.6%, mostly calcium and phosphorus, soluble in water at the pH 7.0 or in alkaline medium. It was tested by the technique of Ambrose-DeEds (11) by the application of chloroform to the skin of rabbits by means of a cotton-tipped applicator. We used a dose of 10 mg. per kilogram of weight of the flavonoid compound injected subcutaneously 20 minutes before the injection of trypan blue (10 cc.) in the marginal ear vein and the application of chloroform wheal to the skin. The average time of the appearance of the dye for 20 rabbits was 42 minutes 54 seconds as against one minute 57 seconds in the control batch of 10 rabbits. These figures when compared with those obtained by Ambrose-DeEds with rutin (three minutes for control and 15.7 minutes for rutin treated rabbits) seem to indicate a higher biological activity of the citrus flavonoid compound than that of rutin.

EXPERIMENTAL

1. Increased capillary fragility induced by leukotaxine

Menkin (19, 20) found that an inflammatory exudate induces an increase in capillary permeability. This was manifested by the accumulation of trypan blue from the circulation in a cutaneous area previously injected with an exudate. This effect was likewise duplicated by leukotaxine, a nitrogenous substance extracted from inflammatory exudation. The effect of leukotaxine on increased capillary permeability is wholly or in part inhibited by the presence of an extract of the adrenal cortex. Hyman and Chambers (21) also found that adrenal cortical extracts are extremely efficient in reducing capillary permeability in perfused isolated limbs. According to them, the cortical extracts have no dilator or constrictor action and presumably affect the capillary wall directly by decreasing the pore size of the intercellular cement.

TABLE I

Inhibitory effect of the flavonoid compound on induced capillary permeability
Rabbits Nos. 37–46

Rabbits	Treatment	Appearance of dye after injection in		
		15 min.	20 min.	30 min.
R. 37, f.	Flavonoid plus 3 mg. leukotaxine	0	0	traces
R. 38, m.		0	0	0
R. 39, m.		0	0	+
R. 40, m.		0	0	0
R. 41, m.		0	traces	traces
R. 42, m.		0	traces	+
R. 43, m.		0	0	0
R. 44, m.		0	traces	+
Controls				
R. 45, m.	No flavonoid and 3 mg. leukotaxine	+	++	++
R. 46, m.		+	++	+++

N.B. Diet of rabbits was Purina rabbit chow checkers. Ingredients of Purina rabbit chow: Ground oats, corn meal, soybean oil meal, linseed oil meal, corn germ meal, dehydrated alfalfa meal, wheat gray middlings, animal protein factor supplement, riboflavin supplement, D activated plant sterol, 2% defluorinated phosphate, .5% iodized salt.

In our experiments we have studied the effect of the flavonoid compound upon the capillary changes induced by leukotaxine using Menkin's technique for indicating these changes. Rabbits of an average weight of 2 kilograms were given 10 mg./kilo/weight of the flavonoid compound 20 minutes before subcutaneous injection of 3 mg. of leukotaxine. Immediately afterwards 12 cc. of 1% trypan blue in saline solution were introduced in the marginal ear vein. The change in capillary permeability was gauged by the degree of dye accumulation in the various skin areas. Altogether 10 rabbits were used for these experiments. The flavonoid compound was injected subcutaneously 10 mg./kilo/weight, shortly before the injections of trypan blue. In Table I the results are summarized.

It appears that the citrus flavonoid compound decreases capillary permeability induced by leukotaxine. Its effect is similar in this respect to that of adrenal cortical extracts, as compared with the data reported by Menkin.

2. Increased capillary fragility induced by bacterial polysaccharide

The polysaccharide isolated by M. J. Shear from *Serratia marcescens* produces an extensive hemorrhage in the tumors of animals and, when the dose is sufficiently large, can even cause death (22, 23). The striking fact is that normal rats and mice are much less susceptible to the toxic effect of this compound and can survive a much larger dose than can the cancerous animals. Four or five hours after the bacterial polysaccharide is injected into the cancerous animals, profuse hemorrhages are also observed in the adrenal gland. When adrenal cortical extract is employed in combination with polysaccharide, tumor breakdown is delayed (instead of in six to seven hours it might occur in 24 hours) and the effect on the adrenal gland is minimized (24).

For our experiments we used August Rat Carcinoma (Crocker Laboratory, Columbia University), a fast growing tumor. As a rule only tumors averaging in size 8–9 cm^3. were used for experimentation. When a rat bearing such a tumor is given 0.5 mg./100 g./weight of a bacterial polysaccharide (P–25) which we received from Dr. Shear, death occurred in about seven hours preceded by profuse hemorrhage in the tumor. Histological examination of the tumor, performed every

TABLE II

Inhibitory effect of the flavonoid compound on induced capillary fragility of tumors (bacterial polysaccharide)

Rat Nos.	Treatment		Result: death or survival
	P-25*	Flavonoid	
171-A, m.	0.5 mg.	3 mg.	Death in 17 hrs.
171-B, f.	0.5 mg.	3 mg.	Death in 22 hrs. 25 min.
171-C, m.	0.5 mg.	3 mg.	Death in 18 hrs.
171-D, m.	0.5 mg.	3 mg.	Death in 19 hrs. 10 min.
171-E, m.	0.5 mg.	3 mg.	Death in 20 hrs. 40 min.
174-A, f.	0.5 mg.	3 mg.	Death in 19 hrs.
174-B, m.	0.45 mg.	3 mg.	Death in 24 hrs. 30 min.
174-C, m.	0.5 mg.	10 mg.	Death in 36 hrs.
174-D, m.	0.5 mg.	10 mg.	Survived
174-E, m.	0.5 mg.	10 mg.	Death in 52 hrs.
174-F, m.	0.5 mg.	10 mg.	Survived
177-A, f.	0.5 mg.	10 mg.	Survived
177-B, m.	0.5 mg.	10 mg.	Death in 66 hrs.
177-C, f.	0.5 mg.	10 mg.	Survived
177-D, m.	0.4 mg.	10 mg.	Survived
177-E, m.	0.45 mg.	10 mg.	Survived
177-F, m.	0.5 mg.	10 mg.	Survived
177-H, m.	0.5 mg.	10 mg.	Death in 26 hrs.
178-A, m.	0.5 mg.	10 mg.	Survived
Controls			
178-B, m.	0.5 mg.	None	Death in 6 hrs. 25 min.
178-C, f.	0.5 mg.	None	Death in 7 hrs. 35 min.
178-D, m.	0.5 mg.	None	Death in 9 hrs.
178-E, m.	0.5 mg.	None	Death in 7 hrs. 30 min.
178-F, f.	0.5 mg.	None	Death in 8 hrs. 20 min.
178-H, m.	0.5 mg.	None	Death in 7 hrs. 15 min.

* P-25 is a preparation of Shear bacterial polysaccharide.

TABLE III

Effect of flavonoid compound on survival of irradiated rats

A. Control group of 40 rats (no flavonoid)
Initial average weight: 183 g.

Number of days of survival:	11	12	13	14	15	16	17	18	19	20	21	22	23
Number of rats succumbed:	1	3	3	2	3	3	3	3	2	4	2	1	2 = 32
Mortality rate:	80%												

B. 20 rats given 4 mg. flavonoid for 10 days
Initial average weight: 186 g.

Number of days of survival:	17	18	19	20	21	22	23	24	25
Number of rats succumbed:	1	1	2	0	0	1	1	1	1 = 8
Mortality rate:	40%								

C. 40 rats given 5 mg. flavonoid for 30 days
Initial average weight: 175 g.

Number of days of survival:	18	19	20	21	22	23	24	25	26
Number of rats succumbed:	1	0	0	0	1	0	0	1	1 = 4
Mortality rate:	10%								

N.B. Basal diet of rats was Purina laboratory chow. Ingredients of basal diet: Meat meal, dried skim milk, wheat germ, fish meal, liver meal, dried beet pulp, corn grits, oat middlings, soybean meal, dehydrated alfalfa meal, molasses, animal protein factor supplement, riboflavin supplement, brewer's dried yeast, thiamin, niacin, vitamin A and D feeding oils, D activated plant sterol, 1% steamed bone meal, 0.5% iodized salt, 0.02% manganese sulfate.

hour after the injection of the compound, revealed that the capillary system of the tumor was first to respond to the toxic effect of polysaccharide by steadily increasing bleeding. There seemingly was present a gradual increase in capillary fragility. When a small dose of 3 mg./100 g./weight of the flavonoid compound was given subcutaneously, one or two hours before the injection of polysaccharide, the life of the animal was prolonged up to 18–24 hours and the capillary hemorrhages considerably delayed and less pronounced. A larger dose of the flavonoid, 5 mg./100 g./weight given twice, before and shortly after the injection of P–25, prevented in many instances the death of the animal. The hemorrhages were still present but in a relatively small area and there was a considerable delay in their appearance, in some instances 48 hours after the injection of P–25. In the group of rats receiving 3 mg./100 g./weight there was a partial breakdown of the tumor which was absent, however, in the majority of the cases receiving 10 mg./100 g./weight of the flavonoid compound.

The following Table II summarizes the results of the experiments on 25 rat bearers of tumor (19 males and six females).

There seems to be some similarity in the protective action of the flavonoid against the hemorrhage-producing effect of bacterial polysaccharide in tumor-bearing rats with that of adrenal cortical extracts. The flavonoid compound gives protection not only to the capillary system of the tumor but to the capillary system of the adrenal gland as well. Both cortex and medulla in the animals treated with polysaccharide and the flavonoid appear relatively normal with few residual hemorrhages in zone X of the cortex.

3. Increased capillary fragility induced by ionizing radiation

In our next series of experiments, radiation injury to the capillary wall was investigated. Field and Rekers (16) in their extensive investigation on dogs found that certain flavonoids gave a considerable protection against radiation, and expressed the opinion that vitamin "P" factors affect the vascular system directly. They concluded that: "previous misunderstanding of the nature of vitamin 'P' has arisen from both the failures to recognize that several flavonone analogues possess very similar anti-hemorrhagic activity and that ascorbic acid has the capacity to potentiate activity in other flavonones."

In our investigation 100 rats, 63 males and 37 females, were submitted to a total body, near-lethal dose of radiation. The average weight of the rats was 180 g., ranging from 160 to 205 g. The radiation factors were: 250 kv, with 0.5 mm. Cu and 3.0 mm. Bakelite Filters. Target distance was 27.5 cm. and 210 r/min. dose rate. All rats received 800 r total body X-radiation in a single exposure. Forty rats served as control. The mortality rate in the control group was 80%. Only eight rats survived at the end of three weeks. Of the treated animals 20 were given 4 mg. of the flavonoid compound for 10 days, three days prior

TABLE IV

The protective action of the flavonoid compound against radiation erythema

14 cases

Case no.	Diagnosis	X-ray dosage	Flav. cpd.	Skin reaction
1. F—38	Carcinoma of cervix	3,100 r skin dose to 4 pelvic fields	600 mg.	Minimal skin reaction. Nausea and vomiting.
2. M—44	Carcinoma of gingiva	3,000 r skin dose to 3 fields	300 mg.	No erythema. No nausea or vomiting.
3. F—61	Carcinoma of pleura	2,900 r to 3 thoracic fields 4,000 r to cerv. f.	300 mg.	No erythema. No nausea. Slight tanning.
4. F—32	Carcinoma of gingiva	2,600 r to 2 skin fields; 10,000 gamma	300 mg.	No skin erythema, dry erythema of mucous.
5. F—44	Carcinoma of cervix	4,000 r skin dose to each of 4 pelvic fields	600 mg.	No erythema. Bleeding arrested.
6. F—50	Hepatoma	3,000 r skin dose to each of 4 abd. portals	300 mg.	Mild erythema. No nausea or vomiting.
7. F—70	Carcinoma of cervix	3,000 r to each of 4 skin portals	300 mg.	Slight nausea. No erythema.
8. F—38	Adenocarcinoma of rectum	5,000 r to each of 4 pelvic fields	600 mg.	No nausea. Slight skin reaction.
9. M—56	Metastatic carcinoma cervical node	4,000 r skin dose delivered in 10 days	300 mg.	Slight dry erythema.
10. F—65	Carcinoma of ovary	1,800 r to each of 4 abd. fields	300 mg.	No erythema.
11. F—45	Scirrhous carcinoma of breast	3,400 r skin dose to each of 4 skin portals	600 mg.	Slight tanning. Skin intact.
12. F—55	Carcinoma of cervix	3,000 r skin dose to each of 4 pelvic portals	600 mg.	No erythema.
13. F—70	Carcinoma of cervix	3,100 r skin dose to each of 4 pelvic portals	300 mg.	Mild skin reaction.
14. M—58	Lymphosarcoma of stomach	3,200 r skin dose to each of 3 abdom. portals	300 mg.	No erythema.

to radiation and seven days post radiation. This dose gave a partial protection. The mortality rate was reduced 40%, with 12 rats surviving the exposure. Forty rats were given 5 mg. of the compound for 30 days, five days prior to the exposure and 25 days post radiation, with a total amount of 150 mg. In this group the mortality rate was reduced to 10% with 36 rats surviving. (Table III.)

From these observations it appears that the citrus flavonoid compound gives considerable if not complete protection to rats against a total body, near-lethal dose of radiation.

CLINICAL

Roentgen sickness is a syndrome in which many factors seem to be involved. It was reported that vomiting and nausea can be alleviated by the use of some antibiotics (aureomycin) and the opinion was expressed that these manifestations are partially due to bacterial infections controlled by antibiotic substances (25). In our clinical studies particular attention was paid to radiation erythema and the biological activity of the flavonoid compound was measured in terms of prevention of the erythematous response. The present report covers 92 patients submitted to large doses of radiation and receiving the flavonoid compound. The preparation was administered orally, 300–500 mg./day for five days prior to the exposure and during the whole course of radiotherapy. Table IV presents the results of the investigation conducted by Dr. Isidore Arons, Department of Radiotherapy, Harlem City Hospital, New York City.

From this preliminary clinical report it appears that the flavonoid compound prevents to a considerable degree the appearance of radiation erythema, while nausea and vomiting might persist in spite of flavonoid therapy.

DISCUSSION

All the available evidence indicates that the capillary wall in normal tissue is comparatively impermeable both to serum albumin and to serum globulin. Consequently the average effective pore size of the capillary wall must be less than six millimicrons. It has been suggested that pore size is confined by the intercellular cement lying between the endothelial cells. It is known from the work of Chambers and Zweifach and others (26, 27) "that the intercellular cement dissolves under certain physical conditions such as the absence of calcium or at slightly acid pH." J. F. Danielli

pointed out that: "It is quite certain that anything which affects the physical condition of the intercellular cement affects the pore size" (28). This in turn will affect the effectiveness of the pores as "perfect sieves" and consequently induce what we know as increased capillary fragility. According to Danielli "under normal conditions associated with increased capillary permeability the walls of the pores through this cement are coated with a layer of adsorbed serum protein which effectively reduces the area of pore through which filtration may occur." Displacement of this adsorbed serum protein by other molecules of small diameter may result in an increase in pore size and in a consequent injury to the capillary wall.

This basic concept of capillary pathology indicates the necessity of making a clear distinction between increased capillary permeability and capillary fragility. The latter term should be used only in those cases where chemical lesions in the capillary wall, or more specifically in the intercellular cement, are present.

As Haden, Schneider and Underwood have shown (29) capillary injury "is by far the most frequent cause of clinical hemorrhagic disease" and this injury "may be due to infection, drugs, toxemia, allergy, or nutritional disturbances." They pointed out that increased capillary fragility is due to changes in the intercellular cement substance or to primary damage to the endothelial cells.

In our present work we have demonstrated that the flavonoid compound gave considerable protection against the toxic effect of leukotaxine, bacterial polysaccharide and ionizing radiation. In all these instances an increase in capillary fragility was prevented by the administration of this compound. Although the mechanism of action of the flavonoid compound is not as yet explained we are inclined to agree with Field and Rekers that vitamin "P" factors affect the capillary system directly, perhaps participating as a principal in the "wear and tear" of a part or all of the capillary system, inhibiting its degeneration and taking part in its regeneration, specifically as far as the intercellular cement is concerned. Seemingly vitamin "P" activity is most evident when chemical lesions in the capillary wall are present and increased capillary fragility exists.

SUMMARY

1. A flavonoid preparation from citrus fruit and containing four identified factors was tested, experimentally and clinically, on its biological activity.

2. Ten mg./kilo/weight of this mixture administered to rabbits 20 minutes before subcutaneous injection of 3 mg. of leukotaxine wholly or in part inhibited the effect of this inflammatory exudate factor on capillary permeability.

3. Ten mg./100 g./weight of this material protected to a considerable degree the capillary system of August Rat Carcinoma and of the adrenal gland of cancerous rat against the toxic effect of 0.5 mg. of bacterial polysaccharide.

4. Five mg. of this preparation administered orally for 30 days, five days prior to the exposure and during 25 days post radiation, gave a considerable but not complete protection against a total body, near-lethal dose of X-ray radiation. The mortality rate in the group of 40 irradiated rats was reduced to 10% as against 80% of mortality in the control batch of 40 irradiated rats.

5. Ninety-two patients submitted to deep X-ray therapy and given the mixed flavonoids for five days prior to the exposure and during the whole period of radiotherapy, 300–600 mg./day, orally, manifested considerable diminution in radiation erythema of the skin and in some cases a complete absence of it. Nausea and vomiting, however, persisted in some of the treated patients.

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CARDIAC OUTPUT IN POSTURAL HYPOTENSION [1]

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The arterial pressure is kept at a nearly constant level in normal man by simultaneous adjustments of the cardiac output and calibre of the arterioles. The normal response on motionless standing is arteriolar constriction and a moderate fall in cardiac output. The arteriolar constriction is usually sufficient to prevent a fall in mean pressure. In certain persons a marked fall in arterial pressure occurs on motionless standing. The purpose of this study is to evaluate the relative importance of an abnormal decline in cardiac output and of a failure of normal vasoconstriction in producing the fall in arterial pressure.

METHODS

Among the 12 patients studied, postural hypotension was idiopathic in three, associated with diabetes mellitus in five, with tabes dorsalis in two, with probable syringomyelia in one, and followed bilateral sympathectomy (T_1 through L_1) for hypertension in one case.[3] All subjects were studied in the morning, usually after a light breakfast. Observations were made on a tilting table in the supine position, and at a foot-down angle which was usually 60° from the horizontal. In the latter position the patient was supported by a foot-plate. The tilted position was maintained for three to four minutes before beginning air collection for determination of oxygen consumption. The air collection usually occupied two minutes, and toward the end of this time blood samples were drawn and pressure measurements were begun. The total duration of tilt was seven to eight minutes. The time allowed for adjustment to the tilted position is therefore quite short. However, if more time is allowed to reach a steady state during the tilt, there is danger of collapse. None of these subjects showed evidence of impending collapse. In one subject (J. D.) A–V blood oxygen differences were determined three minutes apart during the tilt, and in two subjects (C. G. and J. P.) duplicate venous samples were taken approximately one minute apart (before and after drawing the arterial sample). The results, presented in Table I, do not indicate rapid shifts in A–V oxygen difference during the period of measurement.

Albumin infusion consisted of 75 gm. of human serum albumin [4] made up with normal saline to a volume of 1,300 cc. and given intravenously within a period of 15 minutes.

Cardiac outputs were determined by the Fick principle following intracardiac catheterization (1). Mixed venous blood was obtained from the atrium or the pulmonary artery; arterial blood, from the brachial artery through an inlying needle. Blood oxygen contents were determined by a spectrophotometric method (2) or by the method of Van Slyke and Plazin (3). In the latter case, duplicate analyses were required to check within 0.1 vol. %. With the spectrophotometric method, duplicate determinations are not in general done because of the simplicity of the technique. Repeated determinations of arterio-venous oxygen difference on the same blood samples yield results within 0.2 vol. %, unless the hemolyzed samples are grossly turbid. All samples are checked for turbidity before reading. Spectrophotometric oxygen determinations were made in subjects J. D., C. G., C. C., J. P., and C. B. Oxygen consumption was measured by timed collection of expired air in Douglas bags, analysis in the Haldane apparatus, and volume measurements in a Tissot spirometer. Intracardiac and arterial pressures were measured by Hamilton manometers. The point of zero reference was 5 cm. below the fourth costochondral junction in the supine position and in the two instances where measurements were made at −30° (head down). In the upright tilts the zero point was taken at the fourth costochondral junction. Mean arterial pressures were obtained by planimetry.

"Calculated resistance" is expressed as the ratio between mean arterial pressure in mm. Hg and cardiac index in liters per minute per square meter of body surface. This is a general form for the expression of resistance (4). In the present study it is intended to serve as a rough index of over-all changes in the arteriolar system of a given subject. A rise in the function is taken to indicate over-all narrowing of the arteriolar bed, while a fall indicates dilatation. For this purpose it must be admitted that the function is quite imperfect. In the study of regional vascular changes, the general expression for resistance has been refined by corrections for yield pressure and viscosity (5). It is uncertain how much gain in reliability would be achieved by the application of these corrections in the present study. A major uncer-

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[2] U. S. Public Health Service Postdoctorate Research Fellow.

[3] This patient was studied through the courtesy of Dr. Keith Grimson, Department of Surgery, Duke University School of Medicine.

[4] Supplied by the American Red Cross.

tainty arises because comparisons are made of resistances in different body positions. The change from supine to tilted position causes a change in distribution of blood flow to the body, with a resultant change in the contribution of different vascular regions to the total resistance. Since resistance is affected by local factors such as length of small vessels and tissue pressure, and since the influence of these factors is altered by a change in distribution of flow, even the refined formula may not well reflect arteriolar changes in this particular situation. For this reason the general expression is used. Its limitations are emphasized by terming it "calculated resistance." The function is studied only as it changes with body position in a given individual. Direct comparisons of calculated resistance are not made between different subjects, nor are resistances compared in the same subject before and after the administration of albumin.

RESULTS

The data are presented in Table I.[5] The last two cases in the table, G. G. and T. I., usually had

[5] Observations on J. A. were made by Drs. Ebert and Borden of Minneapolis, Minnesota, who kindly made the data available for presentation in this paper.

TABLE I

Subject	Comment	Position	Atrial pressure (mm. Hg)	Arterial pressure (mm. Hg)			Calculated resistance (mm. Hg/L./min./M²)	Blood oxygen content (vol. %)			Oxygen consumption (cc./min./M²)	Cardiac index (L./min./M²)	Heart rate (beats/min.)	Per cent of resting value		
				Systolic	Diastolic	Mean		Arterial	Venous	A-V difference				Mean arterial pressure	Calculated resistance	Cardiac index
J. D.	Female, 65, Idiopathic postural hypotension	Flat	1	164	79	119	44	10.4	6.1	4.3	116	2.7	91			
		50°, 2 min.						11.2	4.9	6.3						
		50°, 5 min.	−5	56	33	43	17	11.7	6.1	5.6	139	2.5	99	36	39	93
		−30°	4	129	65	96	26	12.0	8.7	3.3	121	3.7	93	81	59	137
C. G.	Male, 22, Diabetes, postural hypotension	Flat	0	133	86	107	22	14.5	12.0	2.5	119	4.8	104			
									12.1*							
		60°	−4	86	51	64	19	14.0	9.5	4.5	149	3.3	140	60	86	69
									9.6*							
		−30°	1	139	88	110	22	13.8	10.8	3.0	148	4.9	110	103	100	102
C. C.	Male, 60, Diabetes, postural hypotension	Flat	−4	191	94	130	32	19.1	14.4	4.7	193	4.1	113			
		60°	−6	128	82	96	36	17.8	11.0	6.8	186	2.7	126	74	112	66
J. P.	Male, 43, ?Syringomyelia, postural hypotension	Flat	5	115	72	90	45	19.1	12.1	7.0	138	2.0	57			
									11.6							
		60°	−3	73	55	60	35	19.6	11.2*	8.2		1.7†	101	67	78	85
C. B.	Male, 46, Diabetes, postural hypotension	Flat	0	147	80	111	33	15.1	10.3	4.8	163	3.4	77			
		60°	0	120	76	92	30	15.1	9.8	5.3	162	3.1	103	83	91	91
R. S.	Male, 25, Hypertension, four years post-sympathectomy	Flat		141	82	107	30	19.3	15.1	4.2	151	3.6	47			
		60°		114	71	82	33	19.5	13.8	5.7	141	2.5	75	77	110	70
J. A.	Male, 49, Idiopathic postural hypotension	Flat				77	17	19.3	16.4	2.9	137	4.6				
		70°				63	30	19.6	13.1	6.5	138	2.1		82	176	46
C. H.	Male, 62, Idiopathic postural hypotension	Flat	0	168	83	117	33	16.8	12.5	4.3	151	3.5	71			
		60°	−5	85	56	66	37	17.7	9.0	8.6	154	1.8	94	56	112	51
	Five days later	60°		68	45	53	37	15.4	6.2	9.2	133	1.4	94	45	112	40
	After 75 gm. albumin in 1,300 cc. saline	60°	−5	143	79	108	45	13.0	7.7	5.3	127	2.4	69			
A. B.	Male, 37, Diabetes, postural hypotension	Flat		132	85	103	37	19.7	14.6	5.1	142	2.8	91			
		60°		72	46	57	38	19.9	9.7	10.2	149	1.5	104	55	103	54
	After 75 gm. albumin in 1,300 cc. saline	Flat		167	94	123	23	15.3	12.7	2.6	140	5.4	99			
		60°		140	92	108	43	15.9	10.5	5.4	137	2.5	92	88	187	46
L. M.	Female, 36, Tabes, postural hypotension	Flat	+1	173	117	143	26	12.6	9.6	3.0	163	5.5	103			
		60°	−3	74	45	57	22	13.2	6.9	6.3	167	2.6	118	40	85	47
	After 75 gm. albumin in 1,300 cc. saline	Flat	+8	151	101	125	21	10.6	7.5	3.1	186	6.1	103			
		60°	0	189	127	154	33	11.2	7.5	3.7	173	4.7	144	123	157	77
G. G.	Male, 30, Diabetes, irregular postural hypotension	Flat	−1	99	61	75	17	15.9	12.4	3.5	150	4.3	90			
		60°	−7	85	60	68	34	16.9	7.8	9.1	177	2.0	123	91	200	47
	After 75 gm. albumin in 1,300 cc. saline	Flat	+8	117	70	89	17	13.2	9.8	3.4	177†	5.2	83			
		60°	−2	108	64	80	24	12.9	7.7	5.2	179	3.4	94	91	141	65
	Separate study	Flat	0	115	67	88	19	17.2	13.1	4.1	192	4.7	95			
		60°	−1	107	78	88	42	17.9	9.9	8.0	169	2.1	125	100	220	45
T. I.	Male, 29, Tabes, irregular postural hypotension	Flat	+1	121	72	92	30	18.7	14.7	4.0	123	3.1	103			
		60°	−2	118	80	95	37	18.9	14.1	4.8	123†	2.6	123	103	123	84
	After 75 gm. albumin in 1,300 cc. saline	Flat	+6	155	87	114	37	14.6	10.6	4.0	125	3.1	114			
		60°	+5	151	89	115	34	14.8	11.4	3.4	115	3.4	109	101	92	110

* Samples collected approximately one minute apart.
† Calculation employs preceding oxygen consumption.

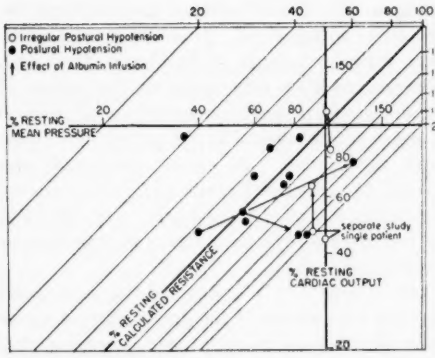

FIG. 1. EFFECT OF CHANGE FROM SUPINE TO UPRIGHT POSITION ON CARDIAC OUTPUT, MEAN ARTERIAL PRESSURE, AND CALCULATED RESISTANCE

The resting, supine state of each subject is represented by the origin. Each plotted point represents a determination in the tilted position. Changes in output, mean arterial pressure, and calculated resistance are expressed as percentage deviations from the resting levels, which are taken to be 100%. Arrows connecting two points indicate that an albumin infusion has been given to a particular subject. The point toward which the arrow is directed represents the effect of tilt after infusion, as referred to measurements in the supine position, also after albumin.

severe weakness and giddiness in the upright position, often had marked postural hypotension, but showed little or none during the study. These patients had no varicosities or other known anatomical abnormalities interfering with venous return. They are classified as having "irregular postural hypotension."

An over-all view of the results is provided by Figure 1. In this figure the resting, supine state of each subject is represented by the origin. Changes in cardiac output, mean arterial pressure, and calculated resistance during tilt are expressed for each subject by the location of a point indicating percentage deviation from the resting levels, which are taken to be 100%. Each plotted point represents a determination in the tilted position. Where two points are connected by an arrow, they both represent the same subject and indicate that an albumin infusion has been given. The point toward which the arrow is directed represents the condition during tilt after albumin has been given. These points, like the others, indicate percentage change from control measurements in the supine position. After the administration of albumin new control measurements were made in the supine position. The values obtained in the subsequent tilt were referred to these new control values.

In Figure 1, the average normal postural reaction would be represented by a region directly below the origin centering about the 75% level on the cardiac output axis. This corresponds to a maintenance of the resting mean arterial pressure, a fall of 25% in cardiac output, and an increase of about 33% in calculated resistance (6, 7).

The two subjects with "irregular postural hypotension" are represented in Figure 1 by open circles. One of these (T. I.) fell within the normal range, since he maintained his pressure and had only a small decline in output on tilt. The other (G. G.), who was studied twice, showed little or no pressure drop on tilt, but had a very large fall in cardiac output. Consequently, this subject had a great increase in calculated resistance during tilt. In terms of maintaining arterial pressure, both of these subjects at the time of study had an adequate vasoconstrictor response to a drop in cardiac output.

The other ten subjects are represented in the figure by solid circles. Eight of these showed little change in calculated resistance during tilt. In very broad terms, the greater the fall in cardiac output, the greater was the drop in arterial pressure, expressed as percentages. In three of these subjects (A. B., C. H., and L. M.) both cardiac output and arterial pressure fell below 60% of the resting levels.

The remaining two subjects show wide deviations from this broad picture. J. A. had a large drop in cardiac output but only an 18% decrease in pressure. There was a large rise in calculated resistance. His behavior is quite like that of G. G., who was classified as "irregular postural hypotension." The difference in classification, which is somewhat artificial, is based on the clear failure of the present subject to maintain his pressure on tilt at the time of study. The other subject, J. D., had the severest postural hypotension of the group. She often fainted in a chair. This patient had the greatest percentage fall in pressure on tilt, but there was little decline in output. There was a large decrease in calculated resistance. When this patient was tilted 30° head down there was a moderate fall in pressure despite an increased cardiac

output. Again, there was a sizable drop in calculated resistance. The other patient (C. G.) who was tilted head down showed no change in output, pressure, or resistance.

In four subjects (C. G., C. C., J. A., and G. G.) the resting cardiac index was between 4.0 and 5.0, and in one (L. M.) it was 5.5. The largest cardiac index falls of the group occurred in three of these subjects during tilt. This suggests that anxiety may have played a part in some of the postural reactions.

Albumin was given to five subjects. In all cases the absolute values of cardiac output and arterial pressure were distinctly higher in the tilted position after albumin than in the tilted position before albumin. In four cases (A. B., L. M., G. G., and T. I.) data are available to describe the changes on tilt after albumin in terms of percentage deviation from the resting level. The results are represented in Figure 1 by the points toward which arrows are directed. The two patients with "irregular" postural hypotension (G. G. and T. I.) continued to maintain their blood pressures during tilt as they had before albumin. As compared with the pre-albumin state, the cardiac outputs were relatively higher and the increase in resistance relatively less. In the other two patients, who had well-marked postural hypotension, the most striking change after albumin was the improvement in maintenance of arterial pressure during tilt. One subject showed a much smaller percentage fall in output than before; the other had a slightly greater percentage fall. Before albumin, neither subject had much change in calculated resistance during tilt. After albumin both cases showed a well-marked increase in calculated resistance during tilt. The increase in calculated resistance was unexpected. It suggests an increase in arteriolar reactivity after albumin, but the expression for resistance is not sufficiently exact to make this secure.

DISCUSSION

In the over-all view, significant postural falls in blood pressure were more consistently associated with failure of normal arteriolar constriction (as indicated by lack of increase in calculated resistance) than with abnormal decline in cardiac output. With a single exception, the subjects with significant postural hypotension showed on tilting a very minor increase in peripheral resistance or an actual fall. This is in general agreement with the findings of Stead and Ebert (8), who concluded on other grounds that failure of arteriolar constriction in response to a fall in blood pressure is fundamental to the production of postural hypotension. A similar conclusion was reached by Ellis and Haynes (9), who measured the cardiac output of two subjects by the acetylene method and found no more than a normal decrease in the tilted position.

One subject (J. D.), who had the most severe postural hypotension of the group, showed a considerable fall in calculated resistance on being tilted in either direction. The mechanism of this fall is not clear. It may result from passive dilatation of arterioles below heart level under increased hydrostatic pressure, thus providing a wide outflow bed. This possibility was suggested by Ellis and Haynes.

The postural fall in cardiac output was variable. From inspection of Figure 1 it is apparent that significant postural hypotension can occur without a greater than normal fall in cardiac output, and that a very large postural fall in cardiac output can occur without a significant drop in blood pressure. However, large pressure drops are often associated with large drops in output. The increase in actual output of tilted subjects after albumin suggests that these large output falls resulted from an inadequate venous return to the heart. The most apparent cause for inadequate return is abnormal dilatation of the venous system under pressure. Filling this system with a larger blood volume would improve venous return to the heart. Normally, the venous system resists passive dilatation under hydrostatic pressure in the upright position, and venous return is well maintained.

Active venous constriction is a familiar response to circulatory inadequacy, as in shock and congestive heart failure. It can occur rapidly in acute pericardial tamponade (10). It is quite possible that an increase in venous tone is an important, normal response to assuming the upright position. The present results suggest that such a response would assist in maintaining the cardiac output, just as arteriolar constriction maintains the blood pressure.

SUMMARY AND CONCLUSIONS

1. Observations have been made on postural reactions of cardiac output and blood pressure in 12 subjects with postural hypotension.

2. Well-marked and even extreme postural hypotension can occur on the basis of failure of arteriolar constriction with no more than a normal postural decrease in cardiac output. Slight to moderate falls in mean arterial pressure have been observed in subjects with very large postural falls in cardiac output and large, though not completely compensatory, increases in calculated resistance. However, large postural falls in arterial pressure were often associated with large falls in cardiac output. In these cases the calculated resistance showed little change on tilt.

3. Large infusions of human serum albumin were given in five cases. In each case there was an increase in cardiac output in the tilted position as compared with the cardiac output in the tilted position before albumin infusion. Arterial pressure was well maintained during tilt. In two cases the response of calculated resistance to posture was greatly increased by the infusion. In two cases, the response was decreased.

4. The data suggest that postural hypotension is primarily dependent upon inadequate arteriolar tone in the upright position. The degree of postural hypotension is often intensified by a large postural fall in cardiac output. It is suggested that this may result from an associated deficiency in venous tone.

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QUANTITATIVE DETERMINATION OF THE VISCERAL PAIN THRESHOLD IN MAN

CHARACTERISTICS OF VISCERAL PAIN, EFFECT OF INFLAMMATION AND ANALGESICS ON THE THRESHOLD, AND RELATIONSHIP OF ANALGESIA TO VISCERAL SPASM

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The development of synthetic analgesics and derivatives of morphine has focused much attention on analgesic testing methods (1). Goetzl, Burrill and Ivy in 1943 abstracted from the literature 75 different methods of quantitative measurement of the pain threshold (2). Pain producing stimuli were divided into mechanical, thermal, chemical and electrical, and all have been used on experimental animals as well as human volunteers. These various stimuli have been applied only to integumental structures, such as skin, lips, teeth and cornea (3, 4). It is generally recognized that visceral pain is a different sensation, elicited only by a special set of stimuli and mediated to the central nervous system through different pathways (5). Little work has been done in the field of visceral algesimetry. Our present evaluation of the effectiveness of various analgesic drugs is based partly on clinical impressions and partly on the assumption that an analysis of response to superficial pain provides an evaluation which also holds true for "deep" or visceral pain (6, 7).

Many of the notorious difficulties of integumental pain threshold determination apply equally to visceral pain. Added to these is the obvious difficulty of inaccessibility of visceral organs. Furthermore, viscera do not respond to the more easily quantitated thermal and electrical stimuli. They respond to only one special mechanical stimulus, that of stretch. From available data it is apparent that visceral pain responses, when obtained, vary a great deal more in intensity, quality and localization than pain responses from integumental structures. In spite of this, a number of valuable investigations has been made concerning localization of pain produced by distention of various portions of the gastro-intestinal tract with balloons.

Quantitative studies of deep analgesia were conducted by Wolff, Hardy and Goodell (6) and Harrison and Bigelow (7) using the method of Lewis, Pickering and Rothschild (8) by which pain is produced by the rhythmic contraction of ischemic muscle of the forearm. From these studies it was concluded that the effects of analgesics on deep pain are similar to those on superficial pain.

The threshold raising effects of narcotics on visceral pain were noted in trained dogs by Yonkman (9). Wolff, Hardy, and Goodell have studied qualitative effects of opiates on visceral pain in man by distending balloons in the duodenum for 40 minutes (6).

The gall bladder and biliary tract, though ordinarily inaccessible, become a fertile region for visceral pain studies after surgical drainage and intubation of any portion of this system. Localization of referred pain from the gall bladder has been studied by distending this organ during cholecystectomy (10, 11), by distending the gall bladder and common bile duct during abdominal operations under local anesthesia or after general anesthesia had worn off (12), and by faradic stimulation of the common bile duct through an electrode incorporated in a common duct catheter (13). Layne and Bergh studied the duration of biliary pain as related to biliary spasm by increasing the intrabiliary pressure with saline solution through a common duct tube (14), and White using the same technique investigated the effect of splanchnicectomy on biliary pain (15).

McGowan and associates observed intrabiliary pain and pressures in patients after choledochostomy by connecting the T-tube in the common bile duct to a manometric system filled with sterile saline solution (16, 17). From these studies they evolved criteria for removal of the T-tube. One of

these criteria was that the "perfusion pain pressure" produced by external elevation of the hydrostatic pressure within the biliary system should be over 500 mm. of water. They felt that pain sensations at lower hydrostatic pressures indicated residual cholangitis. McGowan and the author have utilized these post-operative studies for investigation of a variety of drugs thought to have spasmolytic action on the biliary tract (18-20). Opiates and synthetic analgesics were frequently given during the course of these studies for the production of artificial biliary and duodenal spasm. These drugs were found to raise the "perfusion pain pressure" and the elevation was roughly proportional to the potency and the dosage of the drug. This suggested the use of the T-tube as a pathway to visceral algesimetry.

METHOD

Forty patients volunteered for this study following surgical exploration and intubation of the common bile duct. Of these, 38 also had the gall bladder removed; one patient merely had an exploration and intubation of the common duct and one patient had an exploration of the common duct with a long arm of the T-tube placed through the sphincter of Oddi into the duodenum (Table I).

The original method of McGowan, Butsch and Walters, proposed for intrabiliary pressure recording was employed (16). Two weeks or more after operation, patients were placed in the supine position and the T-tube emerging from the abdomen was connected to a three way manometric system. One limb was connected to an infusion bottle containing sterile saline solution. This permitted filling of the entire system and the biliary ducts with solution. The second limb was connected to a short manometer with a small glass bubble as float arranged to record biliary pressures on a slowly revolving smoked drum. The third limb was connected to a long manometer graduated in mm. which permitted reading of hydrostatic pressures when the infusion bottle was connected to the system and elevated. At times this third water manometer was replaced by a mercury manometer arranged to record pressures on the smoked drum. For all pressure recordings the patients' xyphoid process was chosen arbitrarily as the reference zero point.

The pain threshold was obtained in the following manner: The resting-pressure manometer, being too short for recording of pressures over 250 mm. of water was momentarily cut off from the circuit and the infusion bottle was introduced to the system. The bottle was then slowly raised behind a screen to prevent patients from observing its height. Patients were asked to call out when the first painful sensation was noted. The intrabiliary pressure was recorded at this level by direct reading or by the mercury manometer on the smoked drum. The resting pressure manometer was then substituted for the infusion bottle into the circuit. After each threshold determination, patients were asked to describe the quality, intensity and location of the sensation.

Not all patients tested were suitable subjects for repeated pain threshold recordings. Of the 40 patients who volunteered only 32 could be called upon for further studies. Unsuitable subjects could be divided into three groups. Three patients experienced no sensation of any kind up to pressures of 1,000 mm. of water. Higher pressures were never used. Sufficiently high pressures to cause pain could never be obtained in one patient where the long arm of the T-tube was in the duodenum since infusion fluid flowed into the intestine too rapidly for pressure elevation to develop. In four patients, colicky abdominal pain was easily induced by increased pressures but unlike most patients the pain did not subside together with the fall in intrabiliary pressure. In three patients, colicky pain persisted for five to 10 minutes in spite of normal intrabiliary pressures and medication was required to stop the persistent pain of the fourth patient. These patients were unsuitable for repeated pain level recordings and incidentally only these four patients were unwilling to continue with further experiments.

Resting biliary pressures were recorded for 30 minutes. During this time control pain thresholds were recorded at least three times. These usually checked within 30 mm. of water (Table I). Rapid elevation of the infusion bottle tended to result in slightly higher threshold readings. In later experiments, the speed of pressure elevation was standardized to 30 mm. per second so that even the highest threshold elevations could be obtained in 30 seconds.

After 30 minutes, the analgesic drug to be tested was given by the parenteral route. Pain threshold recordings were made 15 and 30 minutes and one, two and three hours after injection. Occasionally in cooperative patients, thresholds were determined at more frequent intervals. On other occasions experiments were prolonged to four and five hours.

Treatment of data: Both the spontaneous or resting intrabiliary pressure and the pressure sufficient to cause pain were measured in mm. of water with the patients' xyphoid process as the zero reference point. Resting pressure elevations due to the spasmogenic effect of drugs were expressed as elevation in mm. of water since initial resting pressures in patients without biliary sphincter obstruction varied little between 10 and 50 mm. of water (16, 18, 19). Pain threshold elevations due to the analgesic effects of the drugs were measured both in mm. of water and as per cent elevation. The latter form was more useful since initial control pressures necessary to cause pain varied from 90 to 800 mm. of water.

Another value, representing the total analgesic effect of the drug, was obtained by measuring the area under the time-action curve (6). This reflected both the threshold raising activity and the duration of action. The unit for this measure of activity was "per cent-hours."

Peak resting pressures were obtained from the highest point of the kymograph tracings. Peak pain threshold

levels were the highest thresholds measured during a three to four hour period of study. For calculation of the average result of several patients the arithmetic mean was used.

RESULTS

Character and location of pain: Hydrostatic distention of the biliary ducts with pressures up to 800 mm. of water caused some pain sensation in 36 of 40 patients (Table I). The quality and intensity of the pain was described as sharp, stabbing or colicky by 17 patients and was called dull or just an "ache" by 17 other patients. Only two patients noted discomfort which they could not actually call pain with no further increase of this unpleasant sensation up to pressures of 1,000 mm. of water.

The sensation was well localized in 27 patients, usually in the right upper quadrant of the abdomen (23 patients) and occasionally in the epigastric region (4 patients). Five subjects reported pin point localization at a point just below the costal margin in the midclavicular line. The sensation had a more vague distribution in nine patients; it was referred to the entire abdomen by three and to the upper part of the abdomen by six. Some radiation to the back was noted by nine patients; this radiation was never well localized and usually referred to the right upper part of the back. Pain chiefly on the left side over the precordium or radiating down the arm was never reported.

There appeared to be no relationship between the quality and intensity of the preoperative pain and the type of pain sensation reported with these experiments. There was a definite relationship

TABLE I

Location and character of induced biliary pain and its relation to preoperative pain

Case No.	Age	Sex	Duration of intubation	No. of experiments	Postoperative pain threshold	Final pain threshold	Location of preoperative pain	Experimental threshold location	Pain type
1	29	F	3 months	5	440–450	620–630	Epigastrium and back	Epigastric and infrascap.	Sharp
2	58	F	50 days	6	350	500	Right upper quadrant	Right upper quadrant	Ache
3	71	M	3 months	6	700	700–720	Right upper quadrant	Right upper quadrant	Stabbing
4	55	F	2 months	7	620–650	700–710	None, nausea, vomiting	Right upper quadrant	Dull
5	50	F	40 days	7	700	700	Right upper quadrant	Right upper quadrant	Dull
6	23	F	49 days	7	90–100	260–265	Right upper quadrant	Right upper quadrant	Discomf.
7	62	M	26 days	4	400–430	700–720	Generalized abdominal	RUQ and epigastric	Dull
8	40	F	29 days	9	320–330	600	None, vomiting and jaundice	Right subcostal	Sharp
9	32	F	43 days	4	380	510	Entire abdomen	RUQ and infrascapular	Sharp
10	55	M	17 days	4	310–330	400–430	Painless jaundice	Upper abdomen	Dull
11	66	F	22 days	4	450–460	470	Upper abd., rad. to back	RUQ radiating to back	Sharp
12	67	F	22 days	1	—	—	Right upper abdomen	Lasting pain spasm	
13	66	M	63 days	7	260–270	290–300	Upper abdomen	Upper abdomen	Dull
14	67	F	6 months	3	280–300	400	Sharp, rt. upper abdomen	Right upper quadrant	Stabbing
15	53	F	18 days	1	—	500–520	Rt. upper abd., rad. to back	RUQ radiating to back	Dull
16	37	M	68 days	4	160–180	470	Post-gastrectomy jaundice	Right subcostal	Sharp
17	56	M	11 months	1	—	—	Operative ligation of C.D.	T-tube in duodenum	
18	44	M	14 days	1	1,000+	—	Generalized abdominal	No pain to 1,000 mm.	
19	57	F	43 days	8	400–410	490–500	Colicky rt. flank pain	Right upper quadrant	Stabbing
20	64	F	40 days	2	220–230	240–250	Right upper quadrant	Upper abdomen	Discomf.
21	38	M	70 days	1	—	700	Rt. upper abd., rad. to back	Right upper quadrant	Sharp
22	34	F	7 months	7	600–620	700	Jaundice, clay col. stools	Epigastric	Dull
23	53	F	52 days	4	590–600	620–630	Epigastric	Epigastric	Dull
24	41	F	4 months	1	1,000+	—	Epigastric	No pain to 1,000 mm.	
25	39	M	5 months	7	200–210	200–220	Right upper quadrant	Epigastric	Sharp
26	53	F	22 days	2	600–610	600–610	Gen. abd., rad. to back	Upper abd. and back	Dull
27	57	F	42 days	1	—	—	Sharp, rt. upper abdomen	Lasting pain spasm	
28	38	F	24 days	3	700–710	710	Epigastric	RUQ and epigastric	Dull
29	59	F	55 days	1	—	—	Substernal distress	Lasting pain spasms	
30	54	M	14 days	2	800	800	Midabdominal	Upper abdomen	Dull
31	66	F	25 days	7	280–290	300–320	Epigastric	RUQ and epigastric	Sharp
32	29	F	62 days	10	140–150	300	Right upper quadrant	Rt. upper abd. and back	Sharp
33	52	F	18 days	3	600	660–670	Epigastric	Rt. subcostal and epig.	Dull
34	40	M	21 days	3	700	700	Lower abdomen	Right upper quadrant	Sharp
35	43	F	14 days	1	—	—	Epigastric burning	Lasting pain spasms	
36	40	F	26 days	7	550	560–670	Painless, jaundice, nausea	Generalized abdominal	Dull
37	53	F	23 days	7	320–330	400	Upper abdomen	Right upper quadrant	Dull
38	30	F	17 days	7	300	310–320	Rt. upper abd., rad. to back	Rt. upper abd. and back	Sharp
39	41	F	14 days	1	1,000+	—	Generalized abdominal	No pain to 1,000 mm.	
40	36	F	?	1	290–300	—	Nausea, vomiting	Right subcostal	Dull

between the preoperative and postoperative localization of the pain: of six patients who named pain radiating to the back as their chief complaint on entry to the hospital, five later reported back pain as part of the sensation due to distention of the biliary ducts. Chapman and associates have previously reported on this relationship (21). Repeated distention of the biliary system in the same patients resulted in identical localization, intensity and quality of pain both during the course of the same study and on different days. The intensity, quality and localization of the pain was not altered by the rapidity with which the pressure was elevated.

The pain threshold: A great deal of variation was found in the level of the pain threshold from one patient to another. The pain threshold of all patients 14 days after introduction of the T-tube varied from 90 to 800 mm. of water and at the time of tube removal from 200 to 800 mm. of water (Table I). There was less variation of the threshold in the same patients tested at various times. In 15 subjects, mostly those with high initial threshold, the pain level varied no more than 30 mm. during the entire time of intubation, as long as five months in one instance. In the remaining 21 patients, the thresholds slowly increased during the course of observation. A decrease of the threshold was never noted. A slowly increasing threshold over the course of weeks was interpreted as a sign of subsiding cholangitis (16). Sarkisian and McGowan have found a good relationship between the level of the pain threshold and the bacterial count of the bile (22). Similarly inflammation was found to lower the pain threshold during tests of skin pain (23).

In spite of these variations, the biliary pain threshold was found useful as a measure of the effect of analgesic and narcotic substances because variations of threshold in the same patient during the course of a single study were very small (Table I). In 10 subjects repeated threshold determinations on the same day varied by less than 5 mm. of water while in the remaining 24 patients the variation was less than 30 mm. of water, usually no more than 10 mm. In addition, in 15 subjects the threshold varied very little from one day to another often for many weeks and even months (Table I). These patients were well suited for comparative study of the action of several drugs.

TABLE II

Individual threshold determinations of six patients during 12 hours

Time		Patient No.					
Hours	Min.	28	31	32	36	37	38
0	0	700	280	140	560	340	305
0	10	700	280	165	560	360	310
0	20	705	290	145	565	370	310
0	30	710	290	140	565	375	310
1	0	705	290	140	560	370	310
1	30	705	290	140	570	345	320
2	0	710	295	145	570	340	320
2	30	710	290	140	570	350	320
3	0	710	295	145	570	350	320
6	0	695	300	150	555	340	310
6	15	695	295	150	570	350	310
8	0	710	290	150	560	350	320
8	15	710	290	145	570	360	320
11	0	710	310	155	560	375	315
11	15	705	300	150	565	370	320
11	30	705	300	150	570	370	320
12	0	710	295	145	570	370	325
Mean, mm. of water		706	293	147	565	358	316
Standard deviation, mm.		±5.1	±7.1	±5.1	±5.0	±12.6	±5.6
Coefficient of variation, per cent		0.67	2.43	3.82	0.88	3.52	1.77

Near the completion of this study it appeared desirable to obtain data for statistical expression of threshold variations in individual patients. Six patients were studied by repeated threshold determinations over the course of 12 hours (Table II). At intervals these patients returned to the ward for meals and visiting hours and studies were then continued. In all, 17 control readings were obtained. The mean pain thresholds varied from 146.7 to 705.6 mm. of water in these six patients. The smallest standard deviation was ± 5.1 mm. for a mean pressure of 705.6 mm. and the largest standard deviation encountered was ± 12.6 mm. for a mean pressure of 357.9 mm. The smallest coefficient of variation was 0.67 per cent and the largest was 3.82 per cent (Table II). These figures from six selected patients compared favorably with data obtained by the Hardy-Wolff-Goodell technique of individual patients studied from time to time. Uniformity of pain threshold from individual to individual obtained by thermal radiation of the skin could not be duplicated with visceral pain threshold determinations.

The end point: The threshold end point was usually definite particularly in patients who experienced sharp or stabbing pain. A little more care was necessary to determine the exact end point in those patients who experienced merely a dull ache. In the 32 patients found suitable for visceral pain threshold studies the pain sensation

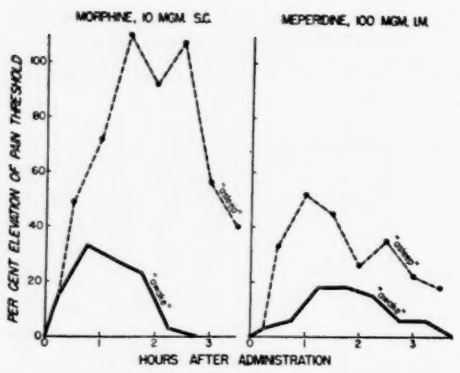

FIG. 1. PERCEPTION OF AND REACTION TO VISCERAL PAIN

For "awake" threshold determinations patients were aroused before the test and asked to signify the first unpleasant sensation due to elevation of the intrabiliary pressure. For the "asleep" curves the pressure was elevated without warning to the inattentive, drowsy or sleeping patients.

subsided almost instantly as the pressure was reduced. In no case was there pain for more than 15 seconds with each threshold determination.

Experiences with pain produced by inflation of balloons in the esophagus, duodenum, colon and ureter suggest that the biliary tree is the most suitable hollow viscus for determination of pain thresholds. Precise hydrostatic measurements of the pain threshold in these other organs were not possible in our experience because these organs responded with peristaltic waves to even slight pressure elevations in the balloon. These contractions were reflected in the pressure readings and steady high pressures within the balloon lumen could therefore not be maintained. Relative to the size of its lumen the common bile duct has by far the thinnest muscular wall of any of the above named organs. Here peristaltic movements are usually not observed during recording of resting pressures. Even with the more sensitive and rapidly responding microphone-condenser electromanometer the only pressure changes recorded were due to changes in abdominal pressure with cardiac systole, respirations and voluntary contractions of the abdominal musculature.

During routine experiments patients were invariably warned of the approaching threshold determination to assure their alertness and then the saline infusion bottle was raised behind the screen. Experiments were also planned during which the patient was notified of the pressure elevation about to occur at the end of 15 and 30 minutes and one, two and three hours and at irregular intervals between these periods the infusion bottle was raised without the patient's knowledge. Figure 1 illustrates two such time action curves of two different patients. Prior to administration of the drug there was no difference between the "awake" and "asleep" thresholds. Fifteen minutes after injection there was a definite elevation of the threshold both after 10 mgm. of morphine and 100 mgm. of meperidine. The two thresholds were still the same. One hour after morphine, there was an "awake" threshold elevation of 30 per cent while the "asleep" threshold elevation was 72 per cent.

TABLE III

Visceral analgesic activity of narcotics and acetylsalicylic acid

Drug	No. of patients	No. of studies	Mean peak threshold elevation, mm. water	Mean peak threshold elevation, per cent	Analgesic activity, "per cent-hours"
Morphine sulfate, 10 mgm., s.c.	30	40	233	56.4	140
Codeine sulfate, 60 mgm., s.c.	22	27	53	11.9	40
Dihydromorphinone hydrochloride, 3 mgm., s.c.	11	11	162	41.9	112
Methyldihydromorphinone hydrochloride, 7.5 mgm., s.c.	7	7	162	46.4	98
Methadone, 7.5 mgm., s.c.	10	10	153	33.2	168
Meperidine hydrochloride, 100 mgm., c.m.	32	50	102	22.9	70
Acetylsalicylic acid, 0.6–1.2 Gm., oral.	12	13	0	0	0

The peak threshold elevations for these two types of determinations were 33 and 110 per cent, respectively, after morphine and 18 and 53 per cent, respectively, after meperidine. Three hours after injection of morphine the "awake" threshold was no longer elevated while the "asleep" threshold was still 40 per cent above the control level.

The first type of threshold, called "awake" was the true perception threshold since the end point was given by alerted patients' first recognition of a painful sensation. The second or "asleep" value was obtained after drowsy, inattentive or even asleep patients were sufficiently stimulated by the pain to make their complaint known. This was therefore not a true threshold but a measure of the analgesic action together with the physiologic and psychologic response to the hypnotic, euphoria producing and other effects of the narcotic. It is apparent that these other effects contribute greatly to the clinical effectiveness of narcotics on visceral pain. Furthermore hypnosis, euphoria and inattentiveness were in evidence for a much longer period of time than the analgesic effect.

Effect of analgesic drugs on the visceral pain threshold: Satisfactory time action curves were obtained from 32 patients on 158 occasions. No ill effects were encountered during or after these studies. Two patients experienced chills about three hours after threshold studies and cholangiography. The temperature, elevated at that time, was found normal three hours later and remained normal thereafter.

The analgesic activity of drugs was compared in two groups of patients. In the first all the patients studied were included (Table III). In the second only seven patients were included whose threshold response was measured for all seven drugs of this series (Figure 2). A true comparison of the effectiveness of the various analgesics could best be made in the latter group since individual initial pain thresholds varied widely and the analgesic effect was dependent to some extent on the magnitude of the initial threshold. These seven patients were among the last ones studied since some of the newer synthetic analgesics had not been available previously; they were patients who showed a good pain threshold end point and whose control thresholds varied little from day to day; lastly they were chosen because the T-tube drainage was continued for a sufficient period of

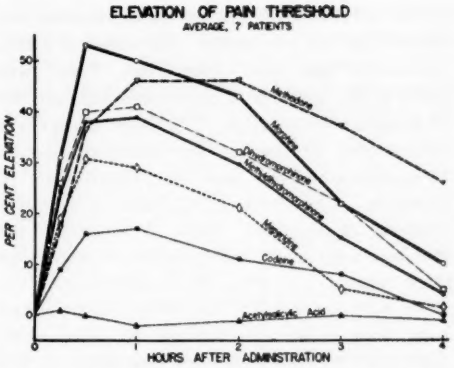

FIG. 2. MEAN TIME-ACTION CURVES IN PER CENT ELEVATION OF PAIN THRESHOLD FOR SEVEN PATIENTS AFTER VARIOUS NARCOTICS AND ACETYLSALICYLIC ACID

time to allow for evaluation of all drugs. Studies were prolonged to four and one-half hours in this group.

Morphine sulfate: Ten mgm. of this agent were given subcutaneously to 30 patients on 40 occasions (Table III). A marked elevation of the visceral pain threshold was observed every time. The onset of action was rapid. The threshold was elevated after 15 minutes in every instance and occasional studies seven minutes after injection showed increase of pressure tolerance at that time. The peak action was observed 15 minutes after injection in 10 per cent of the time-action curves, after 30 minutes in 70 per cent and after one hour in 20 per cent. Some action was still discernible after three hours in all but one patient. The mean peak elevation from the control level was 233 mm. of water or 56.4 per cent and the total analgesic effect was 140 per cent-hours (Table III).

The pain thresholds after morphine of the seven patients in the second group are shown in Table IV. Definite threshold elevation was still present in all patients at the end of four hours. The mean peak elevation of 53 per cent was found 30 minutes after injection.

The mean time-action curves for all drugs for the seven patients of the second group are shown in Figure 2. With the dosage schedule used, morphine caused the highest peak effect and its duration of action was surpassed only by methadone.

Codeine sulfate: This drug was given subcutaneously in 60 mgm. doses to 22 patients on 27 oc-

casions (Table III). A threshold elevation resulted in all but one patient. The onset of action was slower than after other drugs. Only two-thirds of the time-action curves indicated activity 15 minutes after injection. The height of the effect was reached 30 minutes after administration on 10 occasions and one hour after injection on 16 occasions. The duration of action was briefer than that of morphine. Only 72 per cent of the time-action curves showed threshold elevation after three hours. However, all elevations after codeine were so small that the method may well not have been sufficiently delicate to demonstrate slight effects at both ends of the time-action curves. The mean peak elevation for all patients was 53 mm. of water or 11.9 per cent. The total analgesic action of 40 per cent-hours was the smallest encountered.

Comparison of the responses of the seven patients treated with all drugs showed relatively greater variations than those of any other group (Table IV). The mean time-action curve (Figure 2) indicated that codeine caused the smallest threshold elevation of any drug studied. There was no discernible effect at the end of four hours.

Dihydromorphinone hydrochloride ("Dilaudid"): Three mgm. of this drug were given subcutaneously to 11 patients (Table III). The first response, like that after morphine was noted after seven minutes. The peak response was obtained 30 minutes after injection in four patients and one hour after treatment in the remainder. In the

TABLE IV

Visceral pain threshold elevation in mm. of water of seven individual patients after various narcotics

Patient No.	Time after administration													
	Minutes			*Hours*				*Minutes*			*Hours*			
	0	15	30	1	2	3	4	0	15	30	1	2	3	4
	Morphine sulfate, 10 mgm.							Methyldihydromorphinone HCl, 7.5 mgm.						
22	620	640	680	680	670	630	630	200	320	390	420	350	320	250
25	190	360	410	480	390	340	250	300	380	450	450	410	400	330
31	300	400	480	500	460	400	360	300	370	350	350	330	330	330
32	140	300	380	370	340	280	230	560	610	610	610	600	580	550
36	560	620	620	510	600	570	570	320	460	520	530	480	400	330
37	330	400	540	560	540	400	370	310	500	580	460	400	390	370
38	300	480	630	550	500	360	330	600	570	670	780	770	560	540
Mean per cent elevation	0	31	53	50	43	22	10	0	24	38	39	29	15	4
	Codeine sulfate, 60 mgm.							Methadone, 7.5 mgm.						
22	600	620	640	650	620	620	620	700	730	750	770	775	780	740
25	210	260	290	300	240	220	200	210	260	320	380	420	390	360
31	320	360	400	400	380	360	330	290	320	400	480	460	450	390
32	160	200	220	240	190	180	150	150	170	320	360	300	210	190
36	550	550	590	590	580	580	570	550	580	590	610	610	600	570
37	360	430	360	360	370	360	350	310	400	480	500	520	450	430
38	300	300	410	390	390	370	340	300	480	580	570	570	550	510
Mean per cent elevation	0	9	16	17	11	8	2	0	17	37	46	46	37	27
	Dihydromorphinone HCl, 3 mgm.							Meperidine HCl, 100 mgm.						
22	600	620	660	670	630	610	610	600	660	650	650	600	590	600
25	210	340	380	400	320	230	200	200	340	350	340	300	210	210
31	300	430	470	480	460	420	350	340	460	490	470	440	400	375
32	160	280	350	360	340	280	260	210	300	350	340	310	245	225
36	550	575	600	600	560	560	560	550	670	580	590	580	540	540
37	370	420	520	520	520	440	430	375	480	480	470	450	400	380
38	300	460	500	480	450	500	470	300	400	480	470	430	320	320
Mean per cent elevation	0	26	40	41	32	22	6	0	29	31	29	21	5	2

dosages used, dihydromorphinone was superior to methyldihydromorphinone in both peak per cent elevation and total analgesic action. Inspection of individual time-action responses (Table IV) and of their mean curve (Figure 2) revealed fairly uniform action in different patients. The peak of the curve was lower than that of morphine but at three hours their action was identical.

Methyldihydromorphinone hydrochloride[1] (*"Metopon"*): This morphine derivative was given in 7.5 mgm. doses to the seven patients of Table IV. Responses differed little from those obtained with dihydromorphinone. The onset of action was fairly rapid. In one patient the peak was reached in 15 minutes. Of the six others, three showed the maximal response after 30 minutes and three after one hour (Table IV). The shape of the time-action curve was almost identical to that of dihydromorphinone.

Methadone: Ten patients were treated with 7.5 mgm. of this agent (Table III). Although the first analgesic action was noted as soon after injection as after other drugs, the peak activity was definitely delayed. Only one patient showed maximal action after 30 minutes, four showed the peak after one hour, four after two hours and one after three hours. Since methadone was the only drug tested which showed useful activity at the end of four hours threshold determinations were continued beyond four hours in three patients. Useful activity of methadone at five hours was noted in all three and a mean elevation of 8 per cent was still found after six hours. The peak elevation for all ten patients was 153 mm. of water—less than that after morphine and its derivatives. Comparison of the individual (Table IV) and average time-action curves (Figure 2) of seven patients showed the peak action of methadone to be inferior only to morphine. At the end of two hours, methadone was superior to morphine and at three and four hours the activity of this drug was about twice that of morphine and its derivatives.

Meperidine hydrochloride (*"Demerol"*): One-hundred mgm. of this drug were given intramuscularly to all 32 patients on 50 occasions (Table III). It was the most rapidly acting drug of this series. Maximal action was noted after 15 minutes in one-half of the time-action curves while the remainder showed peaks 30 minutes after injection. Meperidine was also the shortest acting drug. Ten curves showed no activity after two hours and one-half of the tests showed no threshold elevation after three hours. The mean peak elevation of 102 mm. of water or 22.9 per cent and the total analgesic action of 70 per cent-hours were the lowest encountered.

Acetylsalicylic acid: Twelve patients were given 0.6 to 1.2 Gm. of acetylsalicylic acid by mouth (Table III). There was no variation from the control thresholds in any instance other than small changes in either direction usually found on repeated control determinations. Sodium salicylate given in comparable doses parenterally to four patients produced no greater changes in the pain threshold than those observed after parenteral administration of sterile saline solution. The composite time-action curve for seven patients (Figure 2) showed changes both above and below the control level which were within the limits of accuracy of the technique.

Relationship of dosage to analgesia: Four patients who were subjected to prolonged biliary drainage and whose control thresholds varied little from day to day received morphine and meperidine in increasing doses.

Time-action curves for one patient who received morphine and another after meperidine are shown in Figure 3. Increases in the lower dosage

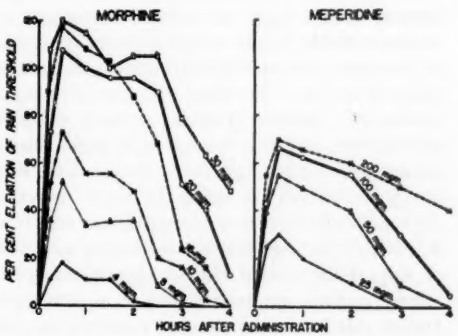

FIG. 3. TIME-ACTION CURVES IN PER CENT ELEVATION OF PAIN THRESHOLD FOLLOWING INCREASING DOSES OF MORPHINE AND MEPERIDINE

[1] Metopon hydrochloride powder for parenteral use was obtained through the kindness of Dr. Nathan B. Eddy, Committee on Drug Addiction and Narcotics of the National Research Council.

TABLE V

Relationship of dosage and visceral analgesia

Morphine sulfate				Meperidine hydrochloride			
Dose, mgm.	Analgesia in per cent-hours	Dose ratio	Analgesia ratio	Dose, mgm.	Analgesia in per cent-hours	Dose ratio	Analgesia ratio
4	22	1	1	25	43	1	1
8	86	2	4	50	114	2	2.7
10	140	2.5	6.4	100	172	4	4.1
16	269	4	12.4	200	239	8	5.6
20	301	5	13.9				
30	329	7.5	15.2				

ranges caused marked elevations of pain threshold. While 4 mgm. of morphine produced a peak threshold increase of 18 per cent, double the amount increased the peak to 52 per cent, and 16 mgm., again double the amount, raised the peak to 118 per cent above the control level. Further increases of the dose to 20 and 30 mgm. caused no higher threshold elevations than 16 mgm. The only effect of the largest doses was a prolongation of the action: no analgesic effect was noted at the end of four hours with doses up to 16 mgm. An appreciable threshold elevation was noted at the end of four hours with 20 mgm. and some action was detected after six hours with 30 mgm.

Time-action curves with increasing doses of meperidine showed a similar pattern of response. Doubling the dose from 25 to 50 mgm. caused a twofold increase of peak threshold elevation. Doubling the dose from 100 to 200 mgm. caused no increase of the peak action but prolonged the action considerably beyond four hours.

Dosages with corresponding per cent-hours and ratio of dosage with ratio of per cent-hours are shown in Table V. Increases in the lower ranges of morphine caused relatively much greater total analgesic action. Doubling the dose of 4 mgm. produced a fourfold increase of total analgesia and doubling the dose from 8 to 16 mgm. tripled the analgesic action. Further increases were relatively ineffective. Doubling 16 mgm. increased the total analgesic action by only 22 per cent from 269 to 329 per cent-hours. Increasing quantities of meperidine caused closely corresponding increases of total analgesic action up to 100 mgm. Double this last amount caused a further increase of analgesia of only 39 per cent. Actual values of the two studies could not be compared since they were done on different patients with different initial pain thresholds. In spite of our limiting pressures to 1,000 mm. of water, it was never possible to obliterate the pain sensation even with the largest doses of morphine.

Relationship of level of initial pain threshold and the analgesic effect: It was shown that in different patients the initial pain threshold varied from 90 to 800 mm. of water (Table I). It soon became apparent that in patients with initially low thresholds a relatively greater rise in pain threshold from analgesic drugs could be obtained than in patients with initially high thresholds. This was true whether the threshold raising action was expressed as elevation in mm. of water or as per cent elevation.

Initial pain thresholds and peak pain thresholds after 10 mgm. of morphine of 40 individual time-action curves are shown in Figure 4. With initial thresholds of 100 to 300 mm. of water, elevations due to morphine by 400 and even 500 mm. were

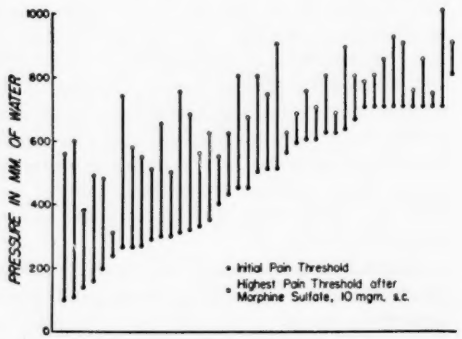

FIG. 4. PEAK THRESHOLD ELEVATIONS IN MM. OF WATER DUE TO 10 MGM. OF MORPHINE

Results of 40 studies arranged in increasing order of initial (control) thresholds.

not uncommon. With the highest initial thresholds of 500 to 700 mm. of water, threshold elevations due to morphine were considerably lower. Many were less than 100 mm. and most not over 200 mm. The mean threshold elevation of nine patients with initial thresholds between 90 and 290 mm. was 340 mm. The mean elevation of 16 patients with initial thresholds of 600 to 800 mm. was 125 mm.

This same phenomenon observed in a single patient who was studied with 100 mgm. meperidine from two to four weeks after operation is illustrated in Figure 5. Two weeks after operation, the initial pain threshold of 320 mm. rose to a peak of 550 mm. after meperidine. This rise of 230 mm. represents a 72 per cent elevation as indicated by the arrow. Further studies 17, 18 and 21 days after operation showed slowly increasing initial pain thresholds and a decreasing effect of the same dose of meperidine. The initial pain threshold 28 days after operation had risen to 600 mm. The increase to 670 mm. after meperidine represented only a 12 per cent elevation of pain threshold.

Patients were further grouped according to their initial pain threshold level and the elevation of threshold due to morphine, 10 mgm., meperidine, 100 mgm., and codeine, 60 mgm., were placed in the various groups (Figure 6). Other analgesics were omitted because of an insufficient number of patients in each group. Here, again, as in individual patients, there was a marked decrease of threshold-elevating activity with control thresholds of increasing magnitude. In the group of patients with control pain thresholds of 90–290 mm., morphine caused a mean rise of pain thresholds of 340 mm., meperidine of 130 mm. and codeine of 100 mm. In the last group with initial thresholds from 700 to 1,000 mm., the three elevations due to morphine, meperidine and codeine were 125, 50 and 40 mm. of water. Narcotics were more effective both relatively and absolutely if the initial visceral pain threshold was low because of inflammation. If the presumption that the early low thresholds are due to cholangitis is correct these findings show a good confirmation of observations made by Schumacher with thermal radiation on inflamed skin (23).

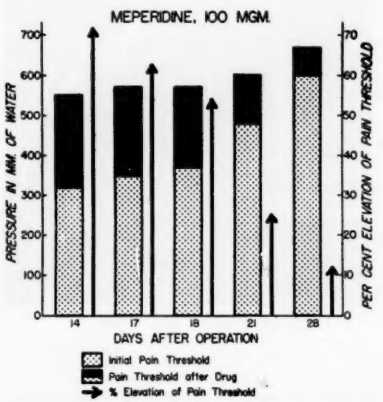
MEPERIDINE, 100 MGM.
PRESSURE IN MM. OF WATER
PER CENT ELEVATION OF PAIN THRESHOLD
DAYS AFTER OPERATION
Initial Pain Threshold
Pain Threshold after Drug
% Elevation of Pain Threshold

FIG. 5. INITIAL PAIN THRESHOLDS AND PEAK THRESHOLDS IN MM. OF WATER AFTER 100 MGM. OF MEPERIDINE OF ONE PATIENT STUDIED AT INTERVALS FROM 14 TO 28 DAYS AFTER OPERATION

Arrows indicate per cent pain threshold elevation due to the medication.

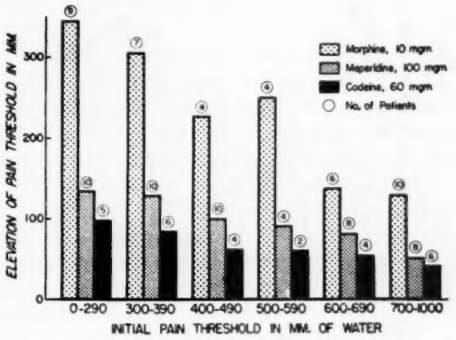

FIG. 6. PEAK PAIN THRESHOLD ELEVATIONS IN MM. OF WATER FOLLOWING MORPHINE, MEPERIDINE AND CODEINE, GROUPED ACCORDING TO MAGNITUDE OF INITIAL (CONTROL) PAIN THRESHOLD

Placebos and the visceral pain threshold: At the onset of these experiments it was suggested that the striking rise of the pain threshold after parenteral injection of analgesic drugs may in part be due to the psychologic effects of the injection. Patients were therefore chosen who had had several previous studies with large doses of potent analgesics. These studies were conducted in the usual manner except that after three initial threshold determinations 1 cc. of sterile saline solution was injected subcutaneously (Figure 7A). A very slight rise of the pain threshold (25 mm. or 8 per cent) was noted after 15 minutes and the

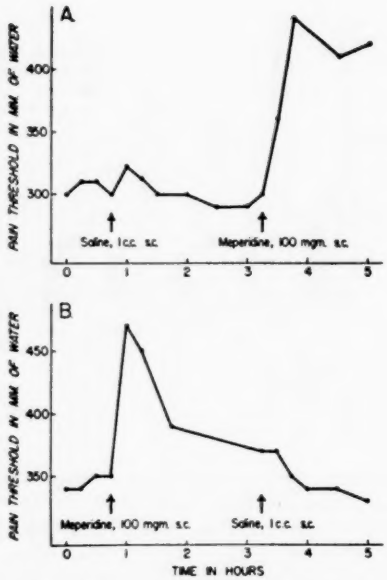

FIG. 7. THE EFFECT OF PARENTERAL PLACEBOS ON THE VISCERAL PAIN THRESHOLD

A. Saline solution followed in two and one half hours by meperidine; B. Meperidine and two and one half hours later saline solution.

threshold then returned to the control level and remained there for three hours. At the end of this time 100 mgm. meperidine were injected with a resultant pressure elevation of 70 mm. (23 per cent) after 15 minutes and 140 mm. (47 per cent) after 30 minutes (Figure 7A). Another patient was given 100 mgm. of meperidine and two and one half hours later, after the effect of the drug had nearly disappeared, 1 cc. of sterile saline solution (Figure 7B). Here there was no increase whatever in the pain threshold after the placebo but a further fall of the threshold as the last traces of the meperidine effect disappeared. From these and six other studies with similar results it was concluded that parenteral administration of placebos did not cause an elevation of the visceral pain threshold. Furthermore, increases of thresholds in suggestible patients were smaller than comparable threshold responses of skin pain (24).

Relationship of visceral analgesia to visceral spasm: The constipating effect of opium was utilized therapeutically many hundred years before its value as a central nervous system depressant was recognized. This effect is common to all the phenanthrene derivatives of opium and is in great part due to lessened motility and increased tone of the gastro-intestinal musculature. This spasmogenic activity on the gastro-intestinal and urinary tracts and the biliary tree constitutes an undesirable side effect in those instances where morphine and codeine are used for their narcotic effect. It was hoped that the appearance of synthetic analgesics would bring a solution to this problem. However, it was shown that both the new synthetic analgesics methadone and meperidine and the morphine derivatives dihydromorphinone and methyldihydromorphinone cause spasm of the small intestine and biliary tree of man (18, 19). The rise of intrabiliary resting pressure which invariably follows the administration of opiates is chiefly due to spasm of the second portion of the duodenum with occlusion of the sphincter of Oddi (16). The rise in pressure has been shown to be proportional to the severity of the spasm (18, 19).

The present method of visceral pain threshold

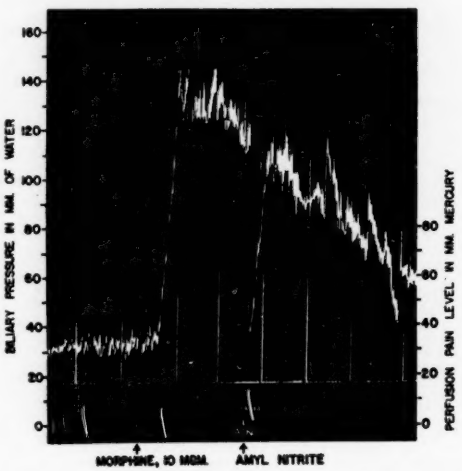

FIG. 8. KYMOGRAPH TRACING OF INTRABILIARY RESTING PRESSURE IN MM. OF WATER AND OF PAIN THRESHOLD IN MM. OF MERCURY FOLLOWING SUBCUTANEOUS INJECTION OF 10 MGM. OF MORPHINE

Three inhalations of amyl nitrite caused a prompt but brief fall in the intrabiliary pressure and had no effect on the pain threshold. The base line (0) is at the level of the xyphoid with the patient in the supine position. Time marker: three minutes.

determination offered a good opportunity for simultaneous quantitative estimation of visceral spasm.

Resting intrabiliary pressures were always recorded during intervals between pain threshold determinations. Occasionally pain threshold levels in mm. of mercury and resting pressures in mm. of water were recorded on the same smoked drum (Figure 8).

The onset of the analgesic effect was always accompanied by a gradual rise of intrabiliary pressure, and the elevation of this pressure was roughly proportional to the elevation of pain threshold (Figure 8). This applied to drugs of varying effectiveness as well as to the same drug given in varying doses. Figure 9 (top) shows the mean peak pain threshold elevations in mm. of water following injection of the six narcotics studied. The same figure (bottom) shows the peak resting pressure elevation after administration of these same drugs. The order of effectiveness of these drugs for both narcosis and spasmogenesis was the same and the ratio of effectiveness of the various agents was similar.

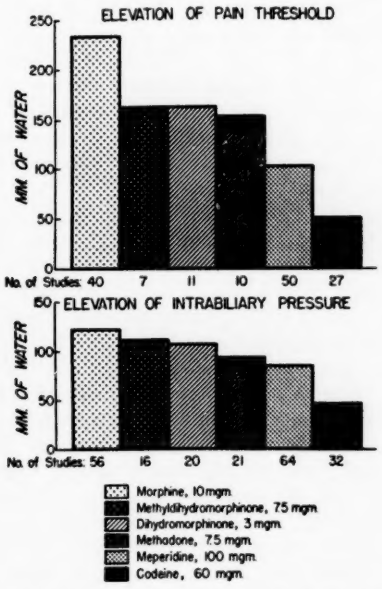

FIG. 9. MEAN PEAK PAIN THRESHOLD ELEVATIONS AND MEAN PEAK INTRABILIARY RESTING PRESSURES IN MM. OF WATER AFTER VARIOUS NARCOTICS

From these data, it is evident that every drug studied which possessed visceral analgesic qualities also caused visceral spasm. The reverse was true also: every drug which was shown to cause biliary and duodenal spasm also had analgesic properties. This is an unexpected and perhaps not significant finding. The analgesic qualities of these agents are due entirely to their action on the central nervous system while their spasmogenic effects are due entirely to local action on smooth muscle (25).

DISCUSSION

Comparison of the effectiveness of various analgesic drugs is difficult because beneficial results depend to such a great extent on the dosage used. Frequently, doses of different agents, necessary to produce the same effect, have been compared. This is misleading since toxic effects of the drugs are thereby disregarded. The "saturation" level, or the highest threshold-raising effect of which a drug is capable (6) provides a good base line for comparison of analgesic effectiveness. This method of comparison was not available to us; first, because the dangers of overdistention of the common bile duct limited us to 1,000 mm. of water pressure, and second, because patient volunteers with common duct drainage, unlike healthy volunteers, are available for a limited time only. It was obvious from the onset that it would not be possible to study the same patients with all drugs and a great variety of dosage forms. We elected, therefore, to study all patients with as many drugs as possible and to confine studies, with a few exceptions, to only one dose and only one method of administration. The dose chosen was the average dose recommended by various sources for a 75 kg. man. The toxic effects of the amounts employed were roughly comparable: in patients without tolerance the dose was one-half of the upper limit of safe usage.

The peak analgesic action for visceral pain occurred earlier than for integumental pain with all drugs tested. The mean peak action for morphine and meperidine at the end of 30 minutes was found about one-half hour earlier than their maximal action determined by the von Frey and Hardy-Wolff-Goodell techniques, (6, 26). The other

agents caused maximal relief of visceral pain 60 minutes after injection whereas their maximal effect on superficial pain was usually found after 90 minutes (26, 27). The measurable duration of action for visceral pain was about half that for skin pain. Wolff and associates found a return to the control threshold after seven hours with 8 mgm. morphine and 6.6 mgm. of methyldihydromorphinone (6). The short duration of action for visceral pain could not be attributed to a lesser overall effect of the drugs because of lower sensitivity of the method since the peak per cent elevation of skin pain due to radiant energy and of visceral pain due to distention were about the same.

Morphine was the most potent analgesic. The morphine derivatives were relatively four times as effective for superficial pain than for deep pain. Equivalent doses based on studies with the Hardy-Wolff-Goodell and von Frey techniques were 30 mgm. morphine for 3 mgm. dihydromorphinone or 6.6 mgm. methyldihydromorphinone (6, 26). The equivalent doses for visceral pain were roughly 8 mgm. of morphine for above doses of the morphine derivatives.

The effect of codeine on visceral pain also compares unfavorably with its action on skin pain. For the latter, the equivalent dose of morphine and codeine was 8:60 (6). For visceral pain this ratio was approximately 3:60.

Since the introduction of meperidine, the evaluation of its analgesic power has been scaled downward somewhat. At present, it is thought to be about twice as effective as codeine and much less effective than morphine (28). This position of meperidine among other analgesics was confirmed. The more rapid onset of action and the shorter duration of action may have been partly due to the fact that meperidine, unlike all other drugs, was injected intramuscularly.

Considerable controversy exists over the effectiveness and duration of action of methadone. Originally it was thought to be twice as effective as morphine both in man and experimental animals (29). Its investigation with the Wolff-Hardy-Goodell technique indicated that 5 mgm. was as potent as 15 mgm. of morphine (28). Clinical experience with methadone in the treatment of visceral pain revealed a somewhat lower potency. Troxil found it 1.5 times as active as morphine (30) and Denton and Beecher found no difference between methadone and morphine, mgm. for mgm., both with respect to speed of action and duration of effect (31). The present study confirmed methadone as a highly active analgesic agent. Present results differ in two respects from previous experience. From observation of patients, it was felt that 7.5 mgm. of methadone caused as much stupefaction, depression and drowsiness as 10 mgm. of morphine. This is in contrast to findings of others who observed no or only minimal sedation and narcosis (28, 29). Secondly, the duration of action of methadone was significantly greater than that of all other drugs tested. The shape of the time-action curve was therefore distinctly different. The total analgesic action of 7.5 mgm. methadone was 20 per cent greater than that of 10 mgm. morphine in spite of the latter's greater peak threshold raising ability.

The total lack of visceral analgesic activity of acetylsalicylic acid was striking. The long established clinical efficacy of acetylsalicylic acid in the treatment of headache, myalgia, arthralgia and other pain originating in integumental structures has been confirmed in the laboratory by quantitative skin threshold studies (32). A 35 per cent elevation of the skin pain threshold due to 0.3 Gm. of acetylsalicylic acid was equal to the effect of 45 mgm. of codeine. Harrison and Bigelow, using the ischemic muscle method, found a 25 per cent threshold elevation after 0.3 Gm. of acetylsalicylic acid (7). They concluded that acetylsalicylic acid is an effective analgesic agent for visceral pain. However, equally high and even higher threshold elevations were obtained with placebos. Furthermore, pain due to muscle ischemia should be grouped with pain from integumental structures and not with visceral pain.

Increasing doses of the same drug showed response patterns similar to those obtained from integumental structures. The "saturation" quantity or the smallest amount with which the highest threshold raising effect was obtained (6) was 16 mgm. for morphine and between 100 and 200 mgm. for meperidine. Both these values were somewhat smaller than those found for skin pain. The "saturation" level, or highest threshold raising effect of which the drug was capable (6) was 120 per cent for morphine and 70 per cent for

meperidine—also somewhat lesser analgesic effects than those observed with skin pain.

SUMMARY AND CONCLUSIONS

1. A quantitative method for measuring visceral pain thresholds by hydrostatic distention of the biliary ducts has been described. The method was used successfully in 32 of 40 patients who volunteered for these studies after surgical exploration of the common bile duct.

2. The painful sensation elicited by external elevation of the intrabiliary pressure was described as sharp or colicky by 47 per cent of the patients. The remainder reported dull pain or an ache. The pain was well localized in the right upper quadrant or epigastric region of the abdomen in 75 per cent of the patients and radiation to the back was noted by 25 per cent. For reliable end point determinations of the threshold the alertness of the patient at that time had to be assured.

3. The initial pain threshold in different patients varied from 90 to 800 mm. of water. A slow rise of pain threshold was often noted in the same patients studied repeatedly from 14 days to seven months after operation. However, all measurements of pain thresholds of the same patients on the same day were found to be at least within ± 10 per cent of their respective average values. In six patients studied at intervals during the course of 12 hours, the control threshold values showed a standard deviation of ± 5.1 to ± 12.6 mm. and a coefficient of variation of 0.67 to 3.82 per cent.

4. The administration of analgesic substances, with the exception of acetylsalicylic acid, invariably resulted in an elevation of the visceral pain threshold. Satisfactory time action curves were obtained on 157 occasions. The action of seven analgesic agents was studied. The doses used were: morphine, 10 mgm.; codeine, 60 mgm.; dihydromorphinone, 3 mgm.; methyldihydromorphinone, 7.5 mgm.; methadone, 7.5 mgm.; meperidine, 100 mgm.; and acetylsalicylic acid, 0.6 Gm.

5. The onset of action, shown by an elevation of the intrabiliary hydrostatic pressure tolerated without pain, was noted between seven and 15 minutes after injection with all drugs studied.

6. The peak threshold elevation was found 30 minutes after administration of morphine and meperidine and 60 minutes after all other agents. The mean peak threshold elevations in per cent together with the number of studies for each drug were as follows: morphine: 40 studies, 56 per cent, codeine: 27 studies, 12 per cent, dihydromorphinone: 11 studies, 42 per cent, methyldihydromorphinone: 7 studies, 46 per cent, methadone: 10 studies, 33 per cent, meperidine: 50 studies, 23 per cent, and acetylsalicylic acid: 13 studies, no threshold elevation.

7. The duration of action was five hours for morphine, four hours for codeine, dihydromorphinone and methyldihydromorphinone, six hours for methadone and three hours for meperidine.

8. The total analgesic action expressed by the area under the time-action curve in "per cent-hours" was 140 for morphine, 40 for codeine, 112 for dihydromorphinone, 98 for methyldihydromorphinone, 168 for methadone, 70 for meperidine and 0 for acetylsalicylic acid.

9. When morphine and meperidine were given in increasing doses, the total analgesic action increased in proportion to the dose up to 16 mgm. for morphine and 100 mgm. for meperidine. A further doubling of the dose to 32 and 200 mgm., respectively, caused only a 23 and 37 per cent respective increase in the total analgesic action.

10. The analgesic effect of various drugs was found to be inversely proportional to the initial pain threshold both in different patients and in the same patients studied at intervals.

11. Parenteral injections of placebos in eight patients were not reflected by a rise in the visceral pain threshold.

12. The severity of biliary and small intestinal spasm due to the analgesic agents studied was found to be in proportion to their analgesic activity.

13. Differences between visceral and integumental pain threshold responses to the various drugs were noted. The peak threshold-raising action for visceral pain occurred earlier than for superficial pain; the duration of action was shorter and the intensity of the analgesia was somewhat smaller for visceral pain. In the doses used, morphine showed the most powerful threshold raising activity and methadone showed the longest action. In contrast to integumental pain, acetylsalicylic acid had no analgesic activity for visceral pain.

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RENAL FUNCTION AND ELECTROLYTE METABOLISM IN ACUTE GLOMERULONEPHRITIS [1]

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Glomerular changes are perhaps the most outstanding anatomical lesions of acute glomerulonephritis. The glomerular tuft usually exhibits increased cellularity with deposits of fibrin or inflammatory exudate in the capsular space. The capsule and basement membranes may become thickened but the arterioles and renal tubules appear relatively normal in the early phases of the disease. Several papers describe a characteristic functional pattern for acute glomerulonephritis which reflects the dominant glomerular lesion (1–4).

The purpose of the present paper is to describe the relation of changes in renal functions to the development of the clinical manifestations of acute glomerulonephritis and of the exacerbation in chronic glomerulonephritis, especially in regard to water and electrolyte metabolism. The renal functions measured include glomerular filtration rate (GFR), effective renal plasma flow (ERPF) and maximum capacity of the renal tubules to excrete p-aminohippurate (Tm_{PAH}) or to reabsorb glucose (Tm_G).

CASE MATERIAL

Facts relative to the diagnosis and onset of the disease for each patient are summarized in Table I. Elevated antistreptolysin titers (5) indicated a recent group A hemolytic streptococcal infection in 10 instances. Evidence of prior renal disease was absent in the eight patients classified as acute glomerulonephritis. Several may have had unrecognized chronic glomerulonephritis and their acute attack may have been an exacerbation rather than the initial attack. However, initial attacks seem likely in the five episodes that "healed" [2] and in the two patients whose urine and blood pressure were normal 16 and 35 days before the onset of signs and symptoms of renal disease. Four instances of exacerbation in chronic glomerulonephritis were observed, one of which occurred in patient 8, whose initial attack of acute glomerulonephritis was also studied. The criterion accepted for the diagnosis of exacerbation in chronic glomerulonephritis was "an abrupt and marked increase in the degree of hematuria" (8). Foreshortening of the latent period between infection and onset of symptoms characteristic of the exacerbation (8, 9) was noted in two of these patients.

The initial and final observations on the urine, blood pressure and edema for each patient are shown in Table II, along with the duration of the abnormalities. The final probable outcomes are indicated in the last column.[2]

The sudden development of nephrotic edema was not accepted as evidence either for the initial acute attack of glomerulonephritis or for an exacerbation in glomerulonephritis. At the time of observation, none of the patients had evidence of the nephrotic phase. Their serum albumin averaged 3.2 gm./100 ml. with a range of 2.5 to 4.5, while their serum cholesterol averaged 219 mg./100 ml. with a range of 179 to 294.

Signs and symptoms of congestive heart failure were present during the acute renal episode in only two patients (patients 9 and 11). These two patients suffered

[1] Supported by a grant from the Life Insurance Medical Research Fund.

[2] Only relatively short follow-up periods were possible in the majority of the patients comprising this study. Thus, certain differentiation between complete healing and the development of chronicity following acute glomerulonephritis was difficult and sometimes impossible (6). However, patients 9, 10 and 11 were known to have chronic glomerulonephritis for one or more years prior to observation. Patient 8 was seen during his first attack and subsequently during an exacerbation. Patients 6 and 7 were seen periodically during one and one half and one years respectively after their initial attacks. During this time proteinuria persisted, a fact which supports the belief that the disease had probably become chronic. Patient 5 was followed for slightly more than one year after his acute glomerulonephritis. The last six urinalyses were normal, quite suggestive of healing. Patient 1 had three normal urines before loss for observation and likewise probably healed. Patients 2, 3 and 4 had three, three and two protein-free urines. However, minimal to moderate microscopic hematuria was still present at the last examination. Since microscopic hematuria may be one of the last abnormal signs to disappear during the healing of acute glomerulonephritis (7), these patients were classified as "probably healing," and included in the "healed" group in the subsequent discussions. Although a follow-up period of a year or more is necessary to differentiate with certainty between healing and chronicity, the presumptive outcomes in this group of patients are designated.

TABLE I

Data concerned with the onset and diagnosis of acute episodes of glomerulonephritis

PATIENT NUMBER	SEX	AGE (years)	PRECEDING INFECTION	MAXIMUM ANTISTREPTOLYSIN TITER (units)	LATENT PERIOD (days)	ONSET NEPHRITIS
1	M	53	Discharge right nostril	100	?	*Acute* Edema eyes, feet Scanty urine Microscopic hematuria Hypertension
2	M	50	Tonsillitis, Fever Rx: Sulfadiazine	2000	20	*Acute* Edema eyes, feet Microscopic hematuria Hypertension
3	M	56	R.U.L. Pneumonia Rx: Penicillin (pneumococci not demonstrated)	700	19	*Acute* (Urine, BP normal 16 days before onset) Edema eyes, feet Microscopic hematuria Hypertension
4	M	35	Severe sore throat	>2000		*Acute* Edema legs Microscopic hematuria Hypertension
5	M	43	Chronic cough "Frequent colds"	300	?	*Acute* Scanty dark urine Edema legs Hypertension
6	M	47	None noted	200	?	*Acute* Abdominal cramps Smoky urine Edema face, legs Hypertension
7	F	46	Postop (cervix) Infection Rx: Penicillin	900	35 (postop)	*Acute* (Urine, BP normal pre-op.) Edema eyes, feet Microscopic hematuria Hypertension
8, #1	M	46	Fever, cough	400	8	*Acute* Bloody urine Edema eyes, feet Hypertension
8, #2			Malaise	500	0	*Exacerbation* Bloody urine Edema eyes, feet Hypertension
9	F	56	Sore throat	---	1	*Exacerbation* (disease known 1 year) Edema feet Flank pain Red urine Hypertension Dyspnea
10	M	63	Cold, fever	400	3	*Exacerbation* (Initial attack 22 years ago) (Exacerbation 12 years ago) Edema legs Pain low back Red urine
11	M	52	None noted	400	?	*Exacerbation* (disease known 15 years) Dyspnea Orthopnea Edema Oliguria

TABLE II

Initial and final observations on urine, edema and blood pressure

PATIENT NUMBER	OBSERVATION	URINE			EDEMA (MAXIMUM WT. LESS USUAL WT.) (lbs)	BLOOD PRESSURE (mm. Hg)	PROBABLE OUTCOME **
		PROTEIN	RBC	CASTS			
1	Admission	+ + + +	10/hpf*	5/hpf	36	180/90	
	Final	0	0	0	0	120/70	Healed
	Duration	(- - - - - - 3½ weeks - - - - - -)			(3 weeks)	(3 weeks)	
2	Admission	+ +	50/hpf	2/hpf	10	210/110	
	Final	0	15/hpf	rare	0	135/80	Healing
	Duration	(- - - - - - 3½ weeks - - - - - -)			(2 weeks)	(2 weeks)	
3	Admission	+ + +	100/hpf	occ.	8	130/100	
	Final	0	10/hpf	occ.	0	110/80	Healing
	Duration	(- - - - - - 6 weeks - - - - - -)			(3 weeks)	(3 weeks)	
4	Admission	+ +	innumerable	occ.	38	198/118	
	Final	0	few	0	0	155/90	Healing
	Duration	(- - - - - - 7 weeks - - - - - -)			(6 weeks)	(9 weeks)	
5	Admission	+	innumerable	rare	21	144/90	
	Final	0	0	0	0	124/70	Healed
	Duration	(- - - - - - 17 weeks - - - - - -)			(5½ weeks)	(10 weeks)	
6	Admission	+ + +	gross	many	33	165/100	
	Final	+	0	0	0	125/85	Chronic
	Duration	(1¼ yrs.+)	(- - 12 weeks - -)		(5 weeks)	(5 weeks)	
7	Admission	+ + +	10/hpf	occ.	22	245/110	
	Final	+ +	0	0	0	130/75	Chronic
	Duration	(persisted)	(- - 10 weeks - -)		(5 weeks)	(9 weeks)	
8, #1	Admission	+ + + +	gross	3/lpf	21	160/110	
	Final	+ + +	many	0	0	130/84	Chronic
	Duration	(- - - - - persisted - - - - - -)			(3 weeks)	(3½ weeks)	
8, #2	Admission	+ + + +	gross	occ.	10	160/100	
	Final	+ +	20/hpf	0	0	130/90	Chronic
	Duration	(- - - - - persisted - - - - - -)			(4 weeks)	(3 weeks)	
9	Admission	+ + +	gross	occ.	18	210/120	
	Final	+ + +	0	0	0	140/80	Chronic
	Duration	(persisted)	(- - 4 weeks - -)		(2½ weeks)	(3 weeks)	
10	Admission	+ + +	gross	many	53	190/110	
	Final	+ +	15/hpf	0	0	180/100	Chronic
	Duration	(- - - - - persisted - - - - - -)			(6 weeks)	(persisted)	
11	Admission	+ + + +	gross	occ.	31	188/120	
	Final	+ +	10/hpf	rare	0	146/84	Chronic
	Duration	(- - - - - persisted - - - - - -)			(10 weeks)	(14 weeks)	

* h.p.f. = high power field

** see footnote 1

from dyspnea and orthopnea, had elevated venous pressures and prolonged circulation time. Their hearts were enlarged.

METHODS

All data presented represent an average of three or more consecutive 15 to 30 minute periods. All observations were performed in the morning. Fluids and food were withheld after the preceding evening meal. Following three periods during which inulin and PAH clearances and electrolyte excretion were observed, Tm glucose or Tm_{PAH} were measured for three consecutive periods. Priming and sustaining infusions containing inulin and PAH were made up in distilled water. The sustaining infusion was delivered by means of a pump at the rate of 1 ml./minute during the first three periods, and at a rate between 2 and 5 ml./minute during Tm measurements. An arterial blood sample was obtained at the midpoint of each urine collection. Plasma, separated from the erythrocytes within 15 minutes of withdrawal of the blood sample, was used for the potassium, chloride, inorganic phosphate, inulin, PAH and glucose determinations. Dried heparin and a small amount of sodium fluoride were used to prevent clotting and glycolysis in the case of glucose and inorganic phosphate, while dried heparin alone was used to prevent clotting in the sample subjected to the other analyses. Serum was used for the sodium determinations.

Inulin was measured by Harrison's (10) modification of the method of Alving, Rubin and Miller (11), PAH by the Bratton-Marshall reaction (12), and glucose by a modification (13) of the Folin method. Sodium in the serum and urine and potassium in the plasma and urine were measured on an internal standard flame photometer. Chlorides were measured by a modification of the Volhard-Harvey method (14), inorganic phosphate by the method of Fiske and SubbaRow (15), and ammonia and urea by Summerson's modification (16) of the Van Slyke and Cullen procedure. Urine pH was determined with the glass electrode.

RESULTS

(*A*) *Renal functions*

Renal functions, weight, blood pressure and urinary abnormalities are recorded in Table III in relation to time of onset and of diuresis. The renal function values are not corrected to a standard surface area because of the complications this would introduce in the subsequent considerations of electrolyte excretions. Glomerular filtration rate was definitely reduced at the first observation in all but one instance (patient 2). The renal plasma flow [3] was less affected and was in the normal range at the first observation in four of the eight initial attacks of acute glomerulonephritis. The pattern of renal dysfunction in both initial attacks and in exacerbations was characteristic of that previously described for acute glomerulonephritis. The filtration fraction (GFR/RPF) was reduced (less than the mean normal value minus two standard deviations) at some time in eight of the 12 episodes. Tm_G was reduced in eight of the nine instances in which it was measured, and Tm_{PAH} in seven of nine instances. The ratios of filtration rate to Tm_G and Tm_{PAH} were usually reduced, indicating a greater impairment of glomerular activity than of tubular function. Several exceptions, however, were noted. In the instance of the four exacerbations the renal functions were undoubtedly abnormal prior to the observations because of the chronic renal disease but exhibited the same general pattern as the initial attacks.

Following the acute episode all functions returned toward normal in patients suffering either their first attack or an exacerbation. Tm_G and Tm_{PAH} were in the normal range at the time of the last observation in four of the five patients who healed or were probably healing (see footnote 2) while the filtration rate had returned to low normal values in three. These findings are similar to those reported in an earlier paper (1) which included observations on five patients suffering their initial attack of glomerulonephritis and two undergoing exacerbations. Serial observations on eight patients in the present study and five patients in the earlier study (1) are available in whom the outcome of the initial attack of acute glomerulonephritis was known (see footnote 2). The first estimation of renal function was obtained within two weeks or less of the onset of the disease in only five of the 13 patients. The serial filtration rates are presented in Figure 1. In general, a smaller reduction in filtration rate and a more rapid return to normal occurred among the six patients whose nephritis healed or probably healed than among the seven whose disease became chronic. Of the nine patients studied within four weeks of the onset of nephritis, the three whose disease failed to heal had initial filtration rates below 55 ml./minute, while those who healed had rates of 44,

[3] The renal plasma flow values in this study are not corrected for any possible incomplete extraction of PAH by the renal tubules. The true renal plasma flow in any instance where there is incomplete extraction of PAH is, therefore, higher than the recorded value.

TABLE III

Blood pressure, urine and renal function tests in patients with acute glomerulonephritis and with exacerbations in chronic glomerulonephritis

PATIENT NUMBER	DAYS AFTER ONSET	RELATION TO DIURESIS	WEIGHT (lbs)	BLOOD PRESSURE (mm. Hg)	URINE PROTEIN	URINE RBC	CONCENTRATION TEST (sp. gr.)	BLOOD UREA (mg/100 ml)	GLOMERULAR FILTRATION RATE (ml/min)	RENAL PLASMA FLOW (ml/min)	FILTRATION FRACTION (percent)	$T_{m_{PAH}}$ (mg/min)	$T_{m_{G}}$ (mg/min)	GF/$T_{m_{PAH}}$	GF/$T_{m_{G}}$
Ave Normal Male (17)															
♂									131	697	19.0	76.1	375	1.72	0.371
									21.5	136	2.4	----	79.7	----	0.0563
1	8	1 day before	184	194/90	3+	10/hpf*	1.012	65.0	82.6	551	15.0	52.2	-----	1.38	------
	16	Last day	158	164/86	0	0	1.012	23.0	78.0	587	13.3	66.1	-----	1.06	------
	30	13 days after	154	120/70	0	0	1.020	----	106	609	17.4	62.1	-----	1.71	------
2	7	First day	151	160/110	2+	20/hpf	1.017	94.7	100	584	17.1	----	377	----	0.27
	21	7 days after	146	130/78	1+	20/hpf	1.017	20.7	102	565	18.1	----	374	----	0.27
3	12	3 days before	151	130/90	2+	50/hpf	1.015	50.1	78.0	369	21.1	----	241	----	0.32
	22	2 days after	144	118/70	1+	10/hpf	1.018	50.3	73.4	369	19.9	57.7	-----	1.27	------
	43	23 days after	148	110/80	0	15/hpf	-----	45.9	73.7	384	19.2	----	199	----	0.37
4	26	1 days before	169	170/110	2+	innumerable	1.011	48.0	43.5	613	7.1	----	184	----	0.24
	30	3rd day during	161	178/110	3+	innumerable	-----	36.3	43.6	481	9.1	18.5	-----	2.38	------
	52	8 days after	136	155/80	0	occ.	1.011	----	47.7	357	13.4	20.1	-----	2.39	------
5	34	4th day during	128	144/90	1+	30/hpf	1.010	30.0	58.9	331	17.8	41.6	-----	1.41	------
	43	4 days after	126	148/86	0	10/hpf	1.011	30.0	64.8	---	----	----	248	----	0.26
	63	24 days after	140	148/86	0	5/hpf	-----	24.0	93.4	---	----	----	333	----	0.28
	118	3 months after	138	124/70	trace	0	1.015	17.4	94.0	526	17.8	----	353	----	0.27
	206	6 months after	150	120/70	0	0	-----	28.4	104	489	21.3	82.0	-----	1.27	------
6	11	3 days before	214	165/95	2+	30/hpf	1.014	90.0	32.6	458	7.1	36.3	-----	0.88	------
	20	7th day during	193	154/90	1+	200/hpf	1.010	61.5	41.4	328	12.6	----	129	----	0.32
	76	5 weeks after	181	125/85	1+	0	-----	26.2	84.8	442	19.2	----	227	----	0.37
7	22	1 day before	177	220/98	3+	5/hpf	1.008	33.7	38.5	282	13.7	----	165	----	0.23
	36	Last day during	146	156/80	2+	0	-----	47.4	37.0	270	13.3	35.8	-----	1.04	------
	54	18 days after	148	130/75	2+	0	-----	56.3	47.7	312	15.3	----	-----	----	------
8, #1	12	2nd day during	138	140/90	4+	30/hpf	1.016	46.6	54.9	523	10.5	----	133	----	0.41
	22	Last day during	127	140/90	4+	innumerable	1.017	49.8	48.7	444	11.0	----	114	----	0.43
	33	11 days after	128	115/80	3+	many	-----	57.2	33.3	234	14.3	15.4	-----	2.16	------
	48	26 days after	126	130/84	3+	occ.	-----	53.4	35.8	260	13.8	----	108	----	0.40
8, #2	12	4 days before	141	176/110	4+	innumerable	1.015	53.0	27.0	117	23.1	6.7	-----	4.03	------
	27	1 day after	131	120/74	3+	25/hpf	-----	45.0	43.4	386	10.7	25.1	-----	1.65	------
9	17	4th day during	147	150/90	2+	4/hpf	1.016	37.8	61.7	356	17.3	----	228	----	0.27
	28	9 days after	147	148/76	3+	0	-----	28.7	74.6	348	21.4	----	269	----	0.28
10	16	3rd day during	182	175/105	3+	innumerable	1.015	40.6	45.0	280	16.1	----	115	----	0.39
	35	22nd day during	156	180/100	3+	15/hpf	-----	33.6	38.6	277	14.0	----	125	----	0.31
11	15	3rd day during	149	176/110	3+	30/hpf	-----	110	21.9	351	6.2	40.8	-----	0.54	------
	40	28th day during	149	144/100	3+	20/hpf	-----	59.6	32.9	394	8.4	41.0	-----	0.80	------
	67	55th day during	131	174/98	2+	10/hpf	-----	27.5	54.4	424	12.8	46.8	-----	1.16	------
	98	22nd day after	136	162/110	2+	occ.	-----	26.7	41.7	220	19.0	40.7	-----	1.02	------
	117	41st day after	135	150/94	2+	40/hpf	-----	33.2	55.5	370	15.0	----	-----	----	------

* h.p.f. = per high power field =

Each renal function datum represents an average of three consecutive periods. Glomerular filtration rates used in calculating GFR to Tm ratios were those measured prior to infusion of large amounts of glucose or PAH. Concentration tests recorded represent the maximum specific gravity attained after a 12 hour restriction of fluid, and were performed within one week of the time indicated.

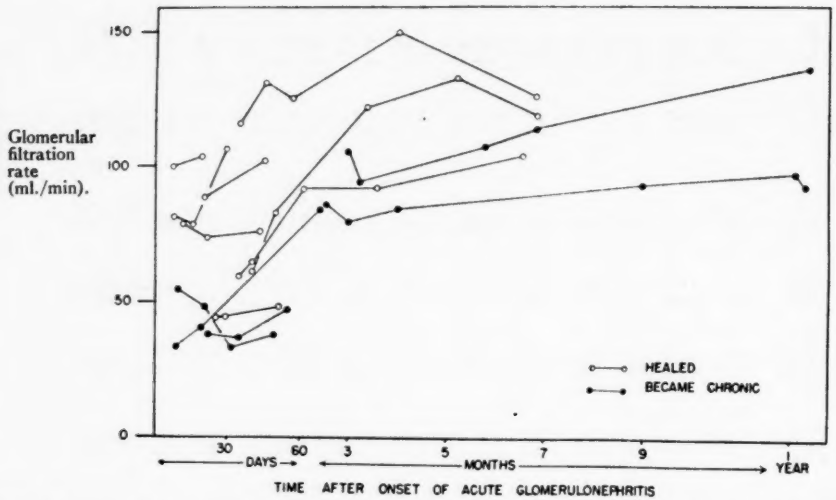


FIG. 1. GLOMERULAR FILTRATION RATE IN RELATION TO TIME AFTER ONSET OF ACUTE GLOMERULONEPHRITIS

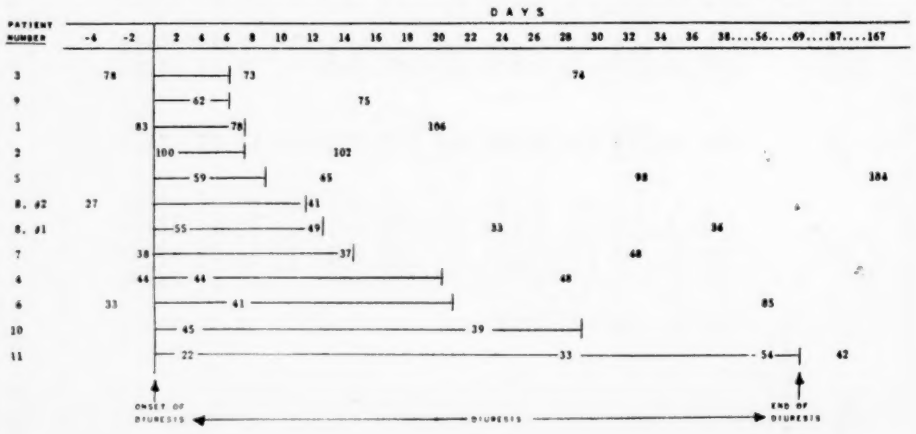
59, 81, 87 and 100 ml./minute. Return of filtration rate to the normal range, however, was not incompatible with the development of chronic renal disease, as in patients 4 and 9 of the earlier study (1). Conversely, patient 6 of the early study and patient 5 of the present study healed completely in spite of rather slow improvement in their glomerular filtration rates. Changes in Tm values showed the same general trends.

Concerning the inter-relations of the several renal functions, the filtration fraction usually increased as the acute renal process improved. However, no correlation could be established between this function or the ratio of filtration rate to excretory or reabsorptive Tm and the eventual outcome.

The exacerbations in chronic glomerulonephritis suffered by patients 9 and 11 were complicated

TABLE IV

Glomerular filtration rate in relation to onset and end of diuresis in acute glomerulonephritis



by congestive heart failure, as evidenced by dyspnea, orthopnea, increased venous pressure, prolonged circulation time and enlargement of the heart. Nevertheless, the functional changes were characteristic of acute glomerulonephritis with a low filtration fraction, rather than that of congestive heart failure with an elevated filtration fraction. Four other patients had slight transient cardiac enlargement by X-ray examination, but no other signs or symptoms suggestive of heart failure. All patients had hypertension at the time of the first measurement of renal function. In five the diastolic blood pressure was 100 mm. Hg or more. In spite of the consistent hypertension, the filtration fraction was greater than the mean normal in only two instances. The blood pressure often returned to normal well before the renal function reached its maximum observed value.

(B) Relation of discrete renal functions to diuresis in the recovery phase of acute glomerulonephritis

The relation of glomerular filtration rate to diuresis in acute episodes of glomerulonephritis is indicated in Table IV. Daily excretion of electrolytes was followed in four of the patients (pa-

TABLE V

The relation of urea clearance to glomerular filtration rate and diuresis in acute glomerulonephritis

PATIENT	RELATION TO DIURESIS	URINE FLOW (ml./min)	GLOMERULAR FILTRATION RATE (GFR) (ml./min.)	INULIN U/P RATIO	UREA CLEARANCE (UC) (ml./min.)	UC/GFR	PLASMA UREA (mg./100 ml)
1	1 day before	1.87	82.6	44.1	35.9	0.44	65.0
	last day during	1.46	78.0	53.5	66.7	0.86	23.0
2	1st day during	6.09	100	16.4	28.0	0.28	94.7
	7 days after	4.07	102	24.5	80.0	0.79	20.7
3	3 days before	2.69	78.0	29.0	42.2	0.54	42.2
	2 days after	1.72	73.4	42.6	39.4	0.54	39.4
	23 days after	2.62	73.7	28.1	43.3	0.59	43.3
4	3 days before	1.30	43.5	33.4	25.7	0.59	48.0
	2 days after	1.75	43.6	25.0	36.3	0.83	36.3
6	7th day during	2.27	41.4	18.2	22.0	0.53	61.5
	5 weeks after	1.92	84.8	44.2	71.8	0.85	26.2
7	1 day before	2.29	38.5	16.8	26.4	0.69	33.7
	last day during	1.24	37.0	29.8	19.5	0.53	47.4
	18 days after	2.21	47.7	21.6	28.6	0.60	56.3
8, #1	2nd day during	1.93	54.9	28.4	35.6	0.65	46.6
	last day during	0.82	48.7	55.5	24.1	0.49	49.8
	11 days after	2.62	33.3	12.7	26.6	0.80	57.2
	26 days after	2.96	35.8	12.1	22.9	0.64	53.4
9	4th day during	0.62	61.7	99.5	16.3	0.27	37.8
	9 days after	1.02	74.6	73.3	31.2	0.42	28.7
10	3rd day during	1.57	45.0	28.7	25.6	0.57	40.6
	22nd day during	1.26	38.6	30.7	26.1	0.68	33.6
11	3rd day during	0.96	21.9	22.8	9.4	0.43	110
	28th day during	0.91	32.9	36.1	15.8	0.48	59.6
	55th day during	3.18	54.4	17.1	32.0	0.59	27.5
	22nd day after	1.57	41.7	26.6	27.5	0.66	26.7
	41st day after	1.57	55.5	36.4	28.5	0.51	33.2

Each datum represents the average of three consecutive periods.

tients 1, 2, 4 and 6). In the remaining patients the weight curve and clinical evidences of edema were utilized to assess diuresis. Filtration rates were measured before the beginning of diuresis in six instances. In the remaining six instances, filtration rate was first observed on the first to fourth day of diuresis (within a few hours of the onset of diuresis in patient 2). The data recorded in Table IV indicate that diuresis following acute glomerulonephritis is not accompanied by a consistent change in filtration rate.

Urea clearance was measured concomitantly with glomerular filtration rate in most of the studies (Table V). The relation of urea clearance to filtration rate and urine flow was not consistent when the group of patients with acute nephritis was considered as a whole. Considerable increases in urea clearance, however, occurred in five patients whose course was characterized by rapid loss of edema in the early phase of diuresis (patients 1, 2, 4, 6 and 9). In three of these the urea clearance increased with no change in glomerular filtration rate, and in each a definite rise in the ratio of urea/inulin ratio occurred. Nor were the increases in urea clearance attributable to concomitant changes in urine flow, which showed only minor variations among the observations obtained for a given patient.

In general, the plasma urea concentration reflected change in urea clearance, and in turn, the filtration rate. Abnormal plasma urea levels, however, did occur in several instances where the filtration rate was not greatly reduced (patients 1–3), while an increasing filtration rate was not incompatible with an increasing plasma urea (patient 7).

In a number of the observations the urea clearance was measured both before and during the infusion of glucose or PAH necessary for the Tm measurements. Urine flow increased in 25 of 27 such observations among 10 patients. The urea clearance increased more than 20 per cent in 11 instances, while the urea/inulin clearance ratio increased in 20 of the 27 observations. In general, the greatest increases were noted among those patients whose urine flows were low during the control periods and who showed substantial diuresis during the osmotic loads. These data indicate that the urea clearance may be influenced by osmotic diuresis in patients with acute glomerulonephritis. The response of these patients to osmotic load was not unlike that observed among patients without renal disease (18).

(*C*) *Effects of acute glomerulonephritis on electrolyte metabolism*

Observations on the plasma or serum levels and the excretion rates of sodium, potassium, chloride, and inorganic phosphate are recorded in Table VI. Data on the excretion of ammonia and urinary pH, and for reference purposes, concurrent filtration rates and urine flows, are included. The fraction of electrolyte filtered at the glomeruli that appeared in the urine also was calculated.

(*1*) *Sodium:* Serum sodium levels were usually in the normal range, although values between 132 and 135 meq./L were observed on occasion in six of the 12 acute episodes of glomerulonephritis. No correlation between the serum sodium level and the presence or absence of edema was noted. Sodium excretion, measured during the determination of filtration rate, was generally greater during the phase of diuresis than at other times. However, the necessity of giving intravenous infusions for clearance purposes probably distorted sodium excretion and urine flow to some extent, limiting the value of such observations. Variations in body weight or daily sodium excretion more accurately reflected sodium balance. But in any case, significant increase in sodium excretion could occur in the absence of change in filtration rate (patients 1 and 4).

(*2*) *Chlorides:* Plasma chloride levels typically were above normal in the patients with acute glomerulonephritis. The mean initial level for the 12 nephritic episodes was 114, $\sigma = 5.0$ meq./L, ranging up to 118 meq./L, as compared to 108, $\sigma = 3.3$ meq./L for 21 normal subjects observed under comparable conditions (*i.e.*, during concomitant measurement of filtration rate and renal plasma flow). All but one of the nephritic patients had plasma chloride levels exceeding 110 meq./L at some time, while only five of the 21 normal subjects had levels greater than this value and none exceeded 114 meq./L. With one exception, the plasma chloride level did not return toward normal within the period of observation as the patients improved from their acute nephritis. Chloride excretion was usually approximately the same as sodium.

TABLE VI

Electrolytes in acute glomerulonephritis

PATIENT NUMBER	DAYS AFTER ONSET	GLOMERULAR FILTRATION RATE (ml/min)	URINE FLOW (ml/min)	SODIUM		CHLORIDE		POTASSIUM			INORGANIC PHOSPHATE			AMMONIA	
				SERUM (meq/L)	EXCRETION RATE (μ/min)	PLASMA (meq/L)	EXCRETION RATE (μ/min)	PLASMA (meq/L)	EXCRETION RATE (μ/min)	PERCENT EXCRETION*	PLASMA (mg/100 ml)	EXCRETION RATE (mg/min)	PERCENT EXCRETION*	EXCRETION RATE (μ/min)	URINE PH
1	8	82.6	1.87	136[a]	40	112	40	4.9	27	6.6	3.8	0.50	15.7	44.0	5.2
	16	78.0	1.46	137	228[b]	110	197	5.1	127	33.0	3.7	0.58	20.1	10.2	7.7
	30	106	1.26	137	337	112	262	4.7	89	18.4	3.1	0.42	12.7	13.3	5.0
2	7	100	6.09	137[a]	550[b]	117	562	4.7	64	13.8	2.7	0.29	10.5	46.4	6.3
	21	102	4.07	138	208	114	228	4.4	83	18.4	2.4	0.27	10.7	15.7	5.9
3	12	78.0	2.65	134[a]	301	117	286	4.8	76	20.6	3.6	0.70	24.6	21.3	6.5
	22	73.4	1.72	138	215	114	233	4.7	70	20.5	2.7	0.58	29.9	34.8	5.7
	43	73.7	2.62	137	310	115	313	5.3	100	25 6	3.5	0.62	26.6	6.8	6.8
4	26	43.5	1.30	133[b]	78	118	113	4.1	73	27.9	3.6	0.17	10.9	24.0	5.0
	30	43.6	1.75	137	209[b]	114	134	5.7	136	55.0	3.9	0.32	18.9	6.2	5.7
	52	47.7	2.93	134	239	116	290	6.0	128	45.1	3.9	0.47	23.7	3.7	7.0
5	34	58.9	1.70	137[a]	152[b]	105	150	3.8	54	23.3	3.2	0.49	25.5	16.1	6.0
	43	64.8	6.33	132	153	106	278	4.2	43	15.6	3.6	0.45	19.7	27.7	5.7
	63	93.4	7.38	133	233	108	317	4.1	124	32.6	3.8	0.47	12.9	28.6	5.4
	118	94.0	3.77	137	213	104	250	4 2	101	25.3	3.4	1.21	37.9	17.0	4.7
	206	104	9.82	136	289	107	302	4.3	127	28 7	3.2	1.28	40.2	19.0	6.6
6	11	32.6	1.38	136[a]	28	118	33	5 1	63	38.0	4.6	0.74	52.2	32.0	4.8
	20	41.4	2.27	138[a]	192[b]	115	147	4.6	93	51.6	4.5	0.95	50.9	4.7	6.7
	76	84.8	1.92	140	238	104	264	4 4	94	25.1	2.2	0.19	10.1	15.8	5.4
7	22	38.5	2.29	140[a]	167	109	151	4.2	37	23.3	3.4	0.43	33.4	9.9	---
	36	37.0	1.24	136	109[b]	114	64	4 8	54	31.0	3.5	0.43	32.5	9.0	6.3
	54	47.7	2.21	135	98	113	133	4 5	87	40.9	3.4	0.53	32.3	7.2	---
8, #1	12	54.9	1.93	134[a]	253	114	213	6.2	79	23.3	4.0	0.49	22.5	4.6	7.6
	22	48.7	0.82	135	66[b]	111	39	4.7	31	13 5	3.4	0.51	30.7	19.7	---
	33	33.3	2.62	140	212[b]	115	210	4.9	87	50 5	4.0	0.47	35.4	6.0	6.6
	48	35.8	2.96	137	260	116	255	6 3	131	57 7	4.3	0.85	55.1	2.4	7.4
8, #2	12	27.0	1.93	134[a]	150	112	144	5.0	68	48.8	5.4	0.64	44.2	---	---
	27	41.4	1.88	132	246	116	272	5.4	96	42.6	4.4	0.80	43.4	---	---
9	17	61.7	0.62	137[a]	49[b]	112	64	4.7	46	16.0	3.5	0.26	12.0	11.5	5.2
	28	74.6	1.02	136	77	109	77	4.3	53	16.3	2.7	0.71	34.8	9.8	6.5
10	16	45.0	1.57	139[a]	78[b]	113	97	3.3	63	42.4	3.2	0.33	23.0	11.4	6.7
	35	38.6	1.26	139[a]	76[b]	112	76	3.8	41	28.2	3.6	0.42	30.5	9.9	6.2
11	15	21.9	0.96	140[a]	27[b]	118	42	7.3	53	32.4	3.9	0.33	40.1	6.2	5.4
	40	32.9	0.91	137[a]	51[b]	118	39	4.4	50	34.6	3.1	0.32	31.2	11.5	5.4
	67	54.4	3.18	139[a]	278[b]	120	272	5.5	89	29.6	3.0	0.49	30.6	6.8	7.5
	98	41.7	1.57	142	170	118	142	4.6	61	32.0	2.6	0.36	33.8	5.4	7.8
	117	55.5	1.57	138	131	118	182	5.3	92	31 4	2.4	0.34	26.4	4.0	6.1

Each datum represents the average of three consecutive periods.
E = During edema; D = During diuresis.
$\frac{\text{amount excreted}}{\text{amount filtered}} \times 100$.
μ = microequivalents.

(*3*) *Potassium:* The average initial plasma potassium level for the 12 acute episodes of nephritis was 5.0 meq./L, $\sigma = 1.08$, with a range of 3.3 to 7.3 meq./L. In contrast, the mean plasma potassium level, obtained under comparable circumstances (*i.e.*, during measurement of glomerular filtration rate and renal plasma flow) in 21 subjects without renal disease was 4.2 meq./L, $\sigma = 0.29$, with a range of 3.4 to 4.8. Only two of the normals had plasma potassium levels greater than 4.5 meq./L, while 10 of the 12 acute nephritics had values greater than this at one time or another. Patient 6 who had relatively low plasma potassium levels was the only one of the group who suffered vomiting prior to the observations. Abnormally high plasma potassium levels occurred in two patients whose filtration rates were not greatly reduced (patients 1 and 3), while conversely several of the patients with the lowest filtration rates had relatively normal plasma potassium levels. As was the case with plasma chloride, potassium plasma concentration was frequently above normal some days after recovery from acute nephritis.

The absolute values for potassium excretion in the urine showed a distinct tendency to rise as diuresis and recovery from the acute renal disease took place. Normally, less than 20 per cent of the potassium filtered at the glomeruli appears in the urine (18). However, the fraction of filtered potassium which was excreted exceeded this value at some time in 10 of the 12 acute episodes of glomerulonephritis. This phenomenon was most apparent among the patients with the greatest reductions in filtration rate but the relationship was not precise. Variations in excretion occurred independently of the plasma potassium level.

(*4*) *Inorganic phosphate:* The plasma level and urinary excretion of inorganic phosphate appear to be affected in acute glomerulonephritis to a lesser and more variable degree than potassium. The average initial plasma inorganic phosphate level for the 12 episodes was 3.7 mg./100 ml., $\sigma = 0.56$ mg./100 ml., as compared to 3.0 mg./100 ml., $\sigma = 0.47$ mg./100 ml. for 13 normal subjects under similar circumstances (*i.e.*, during measurement of filtration rate). Only three of the nephritics had at any time a plasma inorganic phosphate level greater than the mean normal plus two standard deviations. Plasma levels of potassium and inorganic phosphate did not necessarily parallel one another. Unlike potassium, urinary inorganic phosphate excretion increased with improvement in the renal lesion only occasionally. The fraction of filtered inorganic phosphate which was excreted by 13 normal subjects was quite variable, with a range from 2.3 to 26.5 mg./100 ml., a mean of 13.0 mg./100 ml., $\sigma = 7.5$. Nevertheless, the mean normal value plus two standard deviations was exceeded in nine of the 12 episodes of acute nephritis.

(*5*) *Ammonia excretion:* The mean ammonia excretion rate in nine normal subjects, measured during the estimation of glomerular filtration rate and renal plasma flow, was 13 microeq./minute. One subject's rate was 2.7 microeq./minute, while the rates in the remainder varied between 11.6 and 18.2 microeq./minute. Somewhat increased rates of ammonia excretion, varying between 21.3 and 46.4 microeq./minute, were observed in five patients (patients 1–4 and 6) during the early phases of their acute glomerulonephritis (within two weeks of onset). The excretion rates in these patients subsequently fell to between 3.7 and 15.8 microeq./minute. These changes in ammonia excretion could not be correlated precisely with the urinary pH. Several of the remaining patients occasionally exhibited ammonia excretion greater than the normal group, but generally the rates were in the neighborhood of 10 microeq./minute or less.

DISCUSSION

The pattern of renal dysfunction in acute glomerulonephritis appears to be quite characteristic. Typically the glomerular filtration rate is reduced. Function of the proximal convoluted tubules as measured by the maximum excretory capacity for diodrast (Tm_D) or p-aminohippurate (Tm_{PAH}) may also be reduced, but low filtration rate to Tm ratios reflect the greater impairment of glomerular activity. Reabsorptive capacity for glucose may also be impaired but this function and the excretory capacity of the tubules may be affected to different degrees in the same patient.

That the reduced filtration fraction observed in acute glomerulonephritis represents a greater barrier to glomerular filtration than to the passage of blood through the glomerular apparatus has been suggested in an earlier study (1). It is recognized

that, in fatal instances the glomerular tuft changes may be such as to obstruct the flow of blood. This mechanism may account for the usual autopsy description of "bloodless" glomeruli in such patients.[4] However, in milder forms of the disease, high RPF/Tm ratios indicate relative renal hyperemia, and at times, actual renal hyperemia (1).

The characteristic renal functional disturbances of acute glomerulonephritis can persist in spite of circumstances that are ordinarily associated with quite different patterns of dysfunction. Thus, congestive heart failure (19–21) and hypertension (22) are characterized by considerable reduction in the renal plasma flow and by increased filtration fraction. However, a decreased filtration fraction was observed in one patient whose acute glomerulonephritis was complicated by congestive heart failure, while the other patient with the same complication had a normal filtration fraction. Similarly, low filtration fractions occurred in many of the patients with acute nephritis in spite of significant hypertension.

The majority of instances of acute glomerulonephritis heal completely, especially in the younger age group. Unfortunately, accurate prognosis in the early phases of the disease is practically impossible. Renal function studies during the early stages of acute nephritis are of little or no assistance in determining the prognosis. Although the patients in this study who had the greatest impairment of function did not recover, many observers have commented that even the most severe uremia is not incompatible with complete recovery (7). Conversely, the acute disease may be so mild as to almost escape attention and yet fail to heal (7).

The functional changes during the exacerbation of chronic glomerulonephritis are similar to those of the initial acute episode. However, because renal function is usually already reduced to some degree in patients with the chronic disease, recovery of function following the exacerbation is not ordinarily as complete as it is following the initial acute attack.

Edema is one of the most common manifestations of acute glomerulonephritis. Many observers believe that generalized capillary damage accounts for the edema in the majority of instances. This belief is based chiefly on the common localization of edema about the face and especially about the eyes (23), on the occurrence of edema and hypertension prior to or even without urinary abnormalities (24) and on older observations that edema fluid of acute glomerulonephritis contained relatively high amounts of protein (25). However, obvious accumulation of edema in the loose tissue of the face is not surprising in patients with fluid retention who are confined to bed and who do not have local or general elevation of venous pressure. Nor does the development of edema and hypertension prior to the appearance of urinary abnormalities preclude the possibility of early renal functional involvement. And the most recent observation on the edema fluids of acute nephritis reports the low protein content characteristic of transudates rather than of inflammatory reactions (26).

Congestive heart failure (27) was a contributing factor to the edema of only two of the 12 patients with acute glomerulonephritis in this study while none exhibited the nephrotic syndrome. A renal mechanism for fluid and salt retention in acute nephritis, therefore, should receive serious consideration. The observed reduction of filtration rate may well play some role in the pathogenesis of the edema of acute glomerulonephritis. This is especially likely in those patients whose filtration rates were reduced suddenly and severely. However, at least one patient with massive edema had a filtration rate within the normal range. Further, the urea clearance in acute glomerulonephritis is not infrequently reduced out of proportion to the filtration rate and the degree of oliguria. Reabsorption of urea by the renal tubules appears to represent passive diffusion (28). A possible explanation of the low urea/inulin clearance ratio in these instances of acute glomerulonephritis could be damage to the renal epithelium. Such damage perhaps could permit increased back diffusion of salt as well as of urea. Certainly, the present data indicate, at least, that the diuresis during recovery from an episode of acute glomerulonephritis can take place without a concomitant increase in glomerular filtration rate. The increases in urea clearance with diure-

[4] The amount of blood in the glomerular capillaries in an autopsy specimen does not necessarily reflect the amount of blood present during life. Very few erythrocytes are seen in the glomerular capillaries of animals sacrificed with entirely normal kidneys.

sis were not entirely dependent on the increase in urine flow, and thus may have been the result of reconstitution of the renal epithelium with return of function toward normal.

Abnormalities in the plasma concentration and excretion of sodium, potassium, chloride and inorganic phosphate, although common in acute glomerulonephritis were not well correlated with observed changes in renal functions. An increased fraction of filtered potassium excreted in the urine was one of the most consistent electrolyte abnormalities in acute nephritis. This phenomenon has previously been noted in patients with advanced renal failure (29) and probably represents a fortunate shift in glomerulo-tubular balance that permits maintenance of the plasma potassium level as closely as possible to the normal range in the face of decreased filtered loads of the ion. Whether decreased tubular absorption of potassium or increased tubular excretion, both of which theoretically can occur in chronic renal disease (29, 30), is responsible is not certain.

Ammonia excretion appeared to be increased somewhat during the early phases of acute glomerulonephritis. Although rates of ammonia excretion did not correlate precisely with urinary pH, the increased ammonia in the urine perhaps represented a response to the acidosis that may have been present. Subsequently, ammonia excretion fell below the rates usually observed in normals, perhaps because of damage to the distal renal tubule cells.

SUMMARY AND CONCLUSIONS

1. The typical renal functional disturbances in an acute episode of glomerulonephritis issues from the characteristic glomerular lesion which leads to an excessive reduction in filtration rate and filtration fraction. Excretory and reabsorptive functions, as measured by Tm_{PAH} and Tm glucose, respectively, are less extensively reduced.

2. The changes in renal functions are similar in the exacerbation in chronic glomerulonephritis.

3. The typical changes are noted in spite of hypertension or the development of congestive heart failure, conditions otherwise characterized by an increased filtration fraction.

4. The degree of abnormality and altered interrelations of renal functions are of little assistance in arriving at a prognosis. Considerable improvement in all functions usually follows an acute episode, although the rate of recovery is highly variable. Function also improves following an exacerbation but does not return to normal because of pre-existing impairment effected by chronic disease.

5. Salt and water diuresis during the recovery phase of acute glomerulonephritis may take place without change in glomerular filtration rate. Urea clearance is sometimes reduced out of proportion to the filtration rate in acute glomerulonephritis, and during recovery may increase without change in filtration rate.

6. Serum sodium concentration is normal or slightly low while plasma chloride, potassium and inorganic phosphate levels are sometimes above normal. These changes are not related to the degree of abnormality observed in renal functions.

7. Sodium and chloride excretion more or less parallel one another. Potassium excretion is depressed during the early stages of acute glomerulonephritis and increases during the diuresis of the recovery phase. A greater fraction than normal of filtered potassium and inorganic phosphate appear in the urine in the majority of patients.

8. Ammonia excretion is greater than normal during the early phases of acute glomerulonephritis but subsequently falls below the usual normal range.

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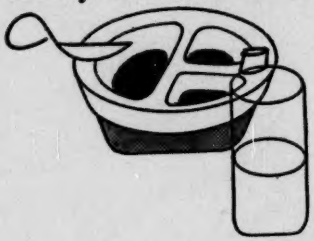

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